Parkinson* & (metal*, manganese, iron, aluminum, cadmium, copper, zinc, mercury, copper) for year 2000-present

Song W, Su H, Song S, Paudel HK, Schipper HM . 2006 Mar. Over-expression of heme oxygenase-1 promotes oxidative mitochondrial damage in rat astroglia. J Cell Physiol 206(3):655-63.
Abstract: Glial heme oxygenase-1 is over-expressed in the CNS of subjects with Alzheimer disease (AD), Parkinson disease (PD) and multiple sclerosis (MS). Up-regulation of HO-1 in rat astroglia has been shown to facilitate iron sequestration by the mitochondrial compartment. To determine whether HO-1 induction promotes mitochondrial oxidative stress, assays for 8-epiPGF(2alpha) (ELISA), protein carbonyls (ELISA) and 8-OHdG (HPLC-EC) were used to quantify oxidative damage to lipids, proteins, and nucleic acids, respectively, in mitochondrial fractions and whole-cell compartments derived from cultured rat astroglia engineered to over-express human (h) HO-1 by transient transfection. Cell viability was assessed by trypan blue exclusion and the MTT assay, and cell proliferation was determined by [(3)H] thymidine incorporation and total cell counts. In rat astrocytes, hHO-1 over-expression (x3 days) resulted in significant oxidative damage to mitochondrial lipids, proteins, and nucleic acids, partial growth arrest, and increased cell death. These effects were attenuated by incubation with 1 microM tin mesoporphyrin, a competitive HO inhibitor, or the iron chelator, deferoxamine. Up-regulation of HO-1 engenders oxidative mitochondrial injury in cultured rat astroglia. Heme-derived ferrous iron and carbon monoxide (CO) may mediate the oxidative modification of mitochondrial lipids, proteins and nucleic acids in these cells. Glial HO-1 hyperactivity may contribute to cellular oxidative stress, pathological iron deposition, and bioenergetic failure characteristic of degenerating and inflamed neural tissues and may constitute a rational target for therapeutic intervention in these conditions. (c) 2005 Wiley-Liss, Inc.

Fored CM, Fryzek JP, Brandt L, Nise G, Sjogren B, McLaughlin JK, Blot WJ, Ekbom A. 2006 Feb. Parkinson's disease and other basal ganglia or movement disorders in a large nationwide cohort of Swedish welders. Occup Environ Med 63(2):135-40.
Abstract: INTRODUCTION: Although it has been hypothesised that metal welding and flame cutting are associated with an increased risk for Parkinson's disease due to manganese released in the welding fume, few rigorous cohort studies have evaluated this risk. METHODS: The authors examined the relation between employment as a welder and all basal ganglia and movement disorders (ICD-10, G20-26) in Sweden using nationwide and population based registers. All men recorded as welders or flame cutters (n = 49,488) in the 1960 or 1970 Swedish National Census were identified and their rates of specific basal ganglia and movement disorders between 1964 and 2003 were compared with those in an age and geographical area matched general population comparison cohort of gainfully employed men (n = 489,572). RESULTS: The overall rate for basal ganglia and movement disorders combined was similar for the welders and flame cutters compared with the general population (adjusted rate ratio (aRR) = 0.91 (95% CI 0.81 to 1.01). Similarly, the rate ratio for PD was 0.89 (95% CI 0.79 to 0.99). Adjusted rate ratios for other individual basal ganglia and movement disorders were also not significantly increased or decreased. Further analyses of Parkinson's disease by attained age, time period of follow up, geographical area of residency, and educational level revealed no significant differences between the welders and the general population. Rates for Parkinson's disease among welders in shipyards, where exposures to welding fumes are higher, were also similar to the general population (aRR = 0.95; 95% CI 0.70 to 1.28). CONCLUSION: This nationwide record linkage study offers no support for a relation between welding and Parkinson's disease or any other specific basal ganglia and movement disorders.

Bates MN. 2006 Jan 27. Mercury amalgam dental fillings: An epidemiologic assessment. Int J Hyg Environ Health .

Abstract: Dental amalgam fillings containing approximately 50% mercury have been used for almost 200 years and have been controversial for almost the same time. Allegations of effects caused by amalgams have involved many diseases. Recent evidence that small amounts of mercury are continuously released from amalgam fillings has fuelled the controversy. This is a comprehensive review of the epidemiologic evidence for the safety of dental amalgam fillings, with an emphasis on methodological issues and identifying gaps in the literature. Studies show little evidence of effects on general chronic disease incidence or mortality. Limited evidence exists for an association with multiple sclerosis, but few studies on either Alzheimer's or Parkinson's diseases. The preponderance of evidence suggests no renal effects and that ill-defined symptom complexes, including chronic fatigue syndrome, are not caused by amalgams. There is little direct evidence that can be used to assess reproductive hazards. Overall, few relevant epidemiologic studies are available. Most prior assessments of possible amalgam health effects have been based on comparisons of dental mercury exposures with occupational exposures causing harm. However, the amalgam-exposed population contains a broader, possibly more susceptible, spectrum of people. Common limitations of population-based studies of dental amalgam effects include inadequate longitudinal exposure assessment and negative confounding by better access to dental care in higher socioeconomic groups. Better designed studies are needed, particularly for investigation of neurodegenerative diseases and effects on infants and children.

Charles LE, Burchfiel CM, Fekedulegn D, Kashon ML, Ross GW, Petrovitch H, Sanderson WT. 2006 Jan 26. Occupational Exposures and Movement Abnormalities among Japanese-American Men: The Honolulu-Asia Aging Study. Neuroepidemiology 26(3):130-139.
Abstract: Objective: The authors analyzed data on 1,049 men aged 71-93 years (excluding those with prevalent Parkinson's disease and stroke) from the Honolulu Heart Program (1965-1968) and the Honolulu-Asia Aging Study (1991-1999) to determine whether occupational exposures to pesticides, solvents, metals, manganese, and mercury during middle age were associated with 14 movement abnormalities 25 years later. Methods: Analyses of variance and multivariate logistic regression were used to assess associations of interest. Results: After adjustment for age, BMI, cognitive functioning, smoking, alcohol drinking, education, and physical activity, there was a positive association between abnormal 'facial expression' and the highest exposure to metals [odds ratio (OR) = 2.62; 95% confidence interval (CI) = 1.35-5.11; trend, p = 0.02], and the highest exposure to mercury (OR = 1.91; 95% CI = 1.04-3.49; trend, p = 0.03). Age was positively associated with all movement abnormalities, and cognitive function, body mass index and physical activity were inversely associated with most movement abnormalities. Conclusion: Higher exposure to any metal, and specifically mercury, was associated with abnormal facial expression. Copyright (c) 2006 S. Karger AG, Basel.

Fasano M, Bergamasco B, Lopiano L. 2006 Jan 17. Modifications of the iron-neuromelanin system in Parkinson's disease. J Neurochem .
Abstract: Parkinson's disease is a common neurodegenerative disorder with a mainly sporadic aetiology, although a number of monogenic familiar forms are known. Most of the motor symptoms are due to selective depletion of dopaminergic, neuromelanin-containing neurones of the substantia nigra pars compacta. Neuromelanin is the dark insoluble macromolecule that confers the black (substantia nigra) or grey (locus coeruleus) colour to monoaminergic basal ganglia. In particular, nigral neurones are pigmented because of the accumulation of by-products of oxidative metabolism of the neurotransmitter dopamine. The occurrence of dopamine (and all the enzymatic machinery required for dopamine synthesis, re-uptake and disposal) and neuromelanin, and a large amount of iron ions that interact with them, makes dopaminergic nigral neurones peculiarly susceptible to oxidative stress conditions that, in turn, may become amplified by the iron-neuromelanin system itself. In this mini-review we describe biophysical evidence for iron-neuromelanin

modifications that support this hypothesis. Furthermore, we discuss the formation of the covalent linkage between alpha-synuclein and neuromelanin from the early stages of the disease.

Bae JH, Jang BC, Suh SI, Ha E, Baik HH, Kim SS, Lee MY, Shin DH. 2006 Jan 14. Manganese induces inducible nitric oxide synthase (iNOS) expression via activation of both MAP kinase and PI3K/Akt pathways in BV2 microglial cells. Neurosci Lett .
Abstract: It is well documented that manganese neurotoxicity induces clinical symptoms similar to those of idiopathic Parkinson's disease. Although microglial cytotoxic mediator-induced neurotoxicity is suggested, the mechanism by which manganese up-regulates cytotoxic mediator, such as nitric oxide (NO), remains poorly understood. Therefore, in this study, we investigated the mechanism of manganese on induction of iNOS in microglial cells. iNOS promoter/luciferase assay revealed that manganese (500(M) regulated the iNOS expression at the transcriptional level. Immunoblot analysis also revealed that phosphorylation levels of ERK, JNK MAPKs and Akt (PKB, PI 3-kinase downstream effector), were increased. Both protein and mRNA levels of iNOS expression were abrogated by specific inhibitors, SP600125 (JNK inhibitor, 20muM), PD98059 (ERKs inhibitor, 50muM), or LY294002 (PI 3-kinase inhibitor, 20muM), but not by SB203580 (20muM), a p38 specific inhibitor. These data lead to the conclusion that manganese regulates the iNOS expression at the transcriptional level in BV2 microglial cells and the increased iNOS protein expression is mediated via both JNK-ERK MAPK and PI3K/Akt signaling pathways, but not via p38 MAPK pathway. Increased iNOS protein level was also found in RAW264.7 murine macrophage cells.

Mattson MP, Meffert MK. 2006 Jan 6. Roles for NF-kappaB in nerve cell survival, plasticity, and disease. Cell Death Differ .
Abstract: Here we review evidence of roles for NF-kappaB in the regulation of developmental and synaptic plasticity, and cell survival in physiological and pathological settings. Signaling pathways modulating NF-kappaB activity include those engaged by neurotrophic factors, neurotransmitters, electrical activity, cytokines, and oxidative stress. Emerging findings support a pivotal role for NF-kappaB as a mediator of transcription-dependent enduring changes in the structure and function of neuronal circuits. Distinct subunits of NF-kappaB may uniquely affect cognition and behavior by regulating specific target genes. NF-kappaB activation can prevent the death of neurons by inducing the production of antiapoptotic proteins such as Bcl-2, IAPs and manganese superoxide dismutase (Mn-SOD). Recent findings indicate that NF-kappaB plays important roles in disorders such as epilepsy, stroke, Alzheimer's and Parkinson's diseases, as well as oncogenesis. Molecular pathways upstream and downstream of NF-kappaB in neurons are being elucidated and may provide novel targets for therapeutic intervention in various neurological disorders.Cell Death and Differentiation advance online publication, 6 January 2006; doi: 10.1038/sj.cdd.4401837.

Wu JH, Xu C, Shan CY, Tan RX. 2006 Jan 2. Antioxidant properties and PC12 cell protective effects of APS-1, a polysaccharide from Aloe vera var. chinensis. Life Sci 78(6):622-30.
Abstract: Through a combination of anion-exchange and repeated gel chromatographies, APS-1 was isolated from fresh leaves of Aloe vera L. var. chinensis (Haw.) Berger (an edible and medicinal plant widely cultivated and consumed in China) as a principal polysaccharide composed of mannose and glucose (ca. 18:5) with its molecular weight around 2.1 x 10(5). In a dose-dependent manner, APS-1 was demonstrated to be free radical scavenging in superoxide and hydroxyl radical assays, inhibitory to the copper-mediated oxidation of human low density lipoprotein (LDL), and protective against hydrogen peroxide (H(2)O(2))-induced lesion to rat PC12 cell (pheochromocytoma cell line). The result suggested that APS-1 could be of considerable preventive and therapeutic significance to some free radical associated health problems such as coronary heart ailments, Parkinson's and Alzheimer's diseases. Furthermore, the finding shed as

well fresh light helpful for a better understanding of the health-benefiting potential of the edible plant consumed by the Chinese people for a couple of centuries.

Youdim MB, Bakhle YS. 2006 Jan. Monoamine oxidase: isoforms and inhibitors in Parkinson's disease and depressive illness. Br J Pharmacol 147 Suppl 1:S287-96.
Abstract: A few years after the foundation of the British Pharmacological Society, monoamine oxidase (MAO) was recognized as an enzyme of crucial interest to pharmacologists because it catalyzed the major inactivation pathway for the catecholamine neurotransmitters, noradrenaline, adrenaline and dopamine (and, later, 5-hydroxytryptamine, as well). Within the next decade, the therapeutic value of inhibitors of MAO in the treatment of depressive illness was established. Although this first clinical use exposed serious side effects, pharmacological interest in, and investigation of, MAO continued, resulting in the characterization of two isoforms, MAO-A and -B, and isoform-selective inhibitors. Selective inhibitors of MAO-B have found a therapeutic role in the treatment of Parkinson's disease and further developments have provided reversible inhibitors of MAO-A, which offer antidepressant activity without the serious side effects of the earlier inhibitors. Clinical observation and subsequent pharmacological analysis have also generated the concept of neuroprotection, reflecting the possibility of slowing, halting and maybe reversing, neurodegeneration in Parkinson's or Alzheimer's diseases. Increased levels of oxidative stress in the brain may be critical for the initiation and progress of neurodegeneration and selective inhibition of brain MAO could contribute importantly to lowering such stress. There are complex interactions between free iron levels in brain and MAO, which may have practical outcomes for depressive disorders. These aspects of MAO and its inhibition and some indication of how this important area of pharmacology and therapeutics might develop in the future are summarized in this review.British Journal of Pharmacology (2006) 147, S287-S296. doi:10.1038/sj.bjp.0706464.

Sung YH, Rospigliosi C, Eliezer D. 2006 Jan. NMR mapping of copper binding sites in alpha-synuclein. Biochim Biophys Acta 1764(1):5-12 .
Abstract: Copper binding to the Parkinson disease-linked protein alpha-synuclein (aS) has been shown to accelerate its oligomerization in vitro and may therefore play a role in aS-mediated pathology in vivo. We use NMR spectroscopy to identify a number of independent copper binding sites in both the lipid-binding N-terminal domain and the highly acidic C-terminal domain of aS. Most of the sites appear to involve negatively charged amino acid side chains, but binding is also observed to the sole histidine residue located at position 50 and to the N-terminal amino group. Both the N-terminal and the histidine sites, as well as the sites in the C-terminal tail, can also bind copper in the more highly structured conformation adopted by aS upon binding to detergent micelles or lipid vesicles. There is no evidence for the formation of any sites requiring long-range order in the protein.

Thompson K, Molina R, Donaghey T, Brain JD, Wessling-Resnick M. 2006. The influence of high iron diet on rat lung manganese absorption. Toxicol Appl Pharmacol 210(1-2):17-23.
Abstract: Individuals chronically exposed to manganese are at high risk for neurotoxic effects of this metal. A primary route of exposure is through respiration, although little is known about pulmonary uptake of metals or factors that modify this process. High dietary iron levels inversely affect intestinal uptake of manganese, and a major goal of this study was to determine if dietary iron loading could increase lung non-heme iron levels and alter manganese absorption. Rats were fed a high iron (1% carbonyl iron) or control diet for 4 weeks. Lung non-heme iron levels increased similar to 2-fold in rats fed the high iron diet. To determine if iron-loading affected manganese uptake, Mn-54 was administered by intratracheal (it) instillation or intravenous (iv) injection for pliarmacokinetic studies. Mn-54 absorption from the lungs to the blood was lower in it-instilled rats fed the

1% carbonyl iron diet. Pharmacokinetics of iv-injected Mn-54 revealed that the isotope was cleared more rapidly from the blood of iron-loaded rats. In situ analysis of divalent metal transporter-1 (DMTI) expression in lung detected mRNA in airway epithelium and bronchus-associated lymphatic tissue (BALT). Staining of the latter was significantly reduced in rats fed the high iron diet. in situ analysis of transferrin receptor (TfR) mRNA showed staining in BALT alone. These data demonstrate that manganese absorption from the lungs to the blood can be modified by iron status and the route of administration. (c) 2005 Elsevier Inc. All rights reserved.

Maharaj DS, Maharaj H, Daya S, Glass BD. 2006. Melatonin and 6-hydroxymelatonin protect against iron-induced neurotoxicity. J Neurochem 96(1):78-81.
Abstract: Oxidative damage of biological macromolecules is a hallmark of most neurodegenerative disorders such as Alzheimer, Parkinson and diffuse Lewy body diseases. Another important phenomenon involved in these disorders is the alteration of iron homeostasis, with an increase in iron levels. The present study investigated whether 6-hydroxymelatonin (6-OHM) can reduce Fe2+-induced lipid peroxidation and necrotic cell damage in the rat hippocampus in vivo. It was found that 6-OHM administration proved successful in reducing Fe2+-induced neurotoxicity in rat hippocampus. This study provides some evidence of the neuroprotective effects of 6-OHM.

Lees-Haley PR, Rohling ML, Langhinrichsen-Rohling J. 2006. A meta-analysis of the neuropsychological effects of occupational exposure to manganese. Clinical Neuropsychologist 20(1):90-107.
Abstract: This article reports a meta-analysis of 25 samples in 20 peer-reviewed published neuropsychological studies of the cognitive, psychological, motor, and sensory/perceptual effects of exposure to manganese. These studies included 1,410 exposed participants and 1,322 controls, for a total N = 2,732. Studies were excluded from this analysis if they were unpublished, had uncodeable data, were based on fewer than four participants, failed to have a comparison group, or reported on manganese effects other than cognitive or sensory/motor motor (e.g., liver functioning). Because the independent variables defining manganese exposure varied across studies, effect sizes were calculated for exposed versus non-exposed workers. Dose-response relations were considered for measures of manganese levels in air/dust (84% of studies reported), blood (MnB; 76% reported), urine (MnU; 52% reported), and hair samples (4% reported). Level of exposure was also estimated by reported years of exposure (M = 13.1 years). Cohen's d statistic yielded a statistically significant weighted mean effect size of -.17, $p < .0001$ for manganese exposure. However, an effect this small is typically undetectable when evaluating individuals because it is smaller (about 1/6 SD) than the confidence intervals of most neuropsychological measures. Because the effect is so slight and the overlap so great between exposed and unexposed participants (87%), the error rate would exceed the hit rate if causal conclusions were rendered for occupational exposure to manganese as the source of an individual's cognitive, sensory, or motor impairments based on neuropsychological testing or symptom reports.

Hazell AS, Normandin L, Norenberg MD, Kennedy G, Yi JH. 2005 Dec 26. Alzheimer type II astrocytic changes following sub-acute exposure to manganese and its prevention by antioxidant treatment. Neurosci Lett .
Abstract: Exposure to manganese in an industrial or clinical setting can lead to manganism, a neurological disorder with similarities to Parkinson's disease. Although the pathogenetic basis of this disorder is unclear, studies indicate this metal is highly accumulated in astrocytes, suggesting an involvement of these glial cells. To investigate this issue, we have used a recently characterized, sub-acute model of manganese neurotoxicity. Treatment of rats with manganese (II) chloride (50mg/kg body weight, i.p.) once daily for 1 or 4 days led to increases in manganese levels of up to 232, 523, and 427% in the cerebral cortex, globus pallidus, and cerebellum, respectively, by instrumental neutron activation analysis.

These changes were accompanied by development of pathological changes in glial morphology identified as Alzheimer type II astrocytosis in both cortical and sub-cortical structures. Co-treatment with either the antioxidant N-acetylcysteine or the manganese chelator 1,2-cyclohexylenedinitrilotetraacetic acid completely blocked this pathology, indicating the cellular transformation may be mediated by oxidative stress associated with the presence of this metal. These findings represent, to our knowledge, the first report of early induction of this pathological hallmark of manganese neurotoxicity, an event previously considered a consequence of chronic exposure to manganese in primates and in human cases of manganism. Our results also indicate that use of this rodent model may provide a novel opportunity to examine the nature and role of the Alzheimer type II astrocyte in the pathophysiology of this disorder as well as in other disease processes in which cerebral accumulation of manganese occurs.

Shamoto-Nagai M, Maruyama W, Yi H, Akao Y, Tribl F, Gerlach M, Osawa T, Riederer P, Naoi M. 2005 Dec 16. Neuromelanin induces oxidative stress in mitochondria through release of iron: mechanism behind the inhibition of 26S proteasome. J Neural Transm .
Abstract: Parkinson's disease is characterized by the selective depletion of dopamine neurons in the substantia nigra, particular those containing neuromelanin. Involvement of neuromelanin in the pathogenesis may be either cytotoxic or protective. Recently we found that neuromelanin reduces the activity of 26S proteasome. In this paper, the detailed mechanisms behind the reduced activity were studied using neuromelanin isolated from the human brain. Neuromelanin increased the oxidative stress, but synthetic melanin did not. Superoxide dismutase and deferoxamine completely suppressed the increase, indicating that superoxide produced by an iron-mediated reaction plays a central role. Iron was shown to reduce in situ 26S proteasome activity in SH-SY5Y cells and the reduction was protected by antioxidants. These results suggest that iron released from neuromelanin increases oxidative stress in mitochondria, and then causes mitochondrial dysfunction and reduces proteasome function. The role of neuromelanin is discussed in relation to the selective vulnerability of dopamine neurons in Parkinson's disease.

Martin CJ. 2005 Dec 3. Manganese neurotoxicity: Connecting the dots along the continuum of dysfunction. Neurotoxicology .
Abstract: Three different manifestations of manganese neurotoxicity have been described. The first, and historically most prominent, is often termed manganism: a dramatic extrapyramidal syndrome following acute, overwhelming exposure. While resembling Idiopathic Parkinson's Disease (IPD), most authorities have regarded the two conditions as clinically and pathophysiologically distinct. The second manifestation, reported by several investigators starting in the 1980s, consisted of subclinical and subfunctional declines in the performance of specialized neuropsychological tests. The implication of these cross-sectional findings was that, when superimposed upon age-related attritional effects, increased rates of clinical disease could result. In this decade, it has been proposed that manganese exposure may play a role in the development of IPD itself. Investigating the relationship between these three manifestations should be a priority for future research.

Park RM, Bowler RM, Eggerth DE, Diamond E, Spencer KJ. 2005 Dec 2. Issues in neurological risk assessment for occupational exposures: The Bay Bridge welders. Neurotoxicology .
Abstract: The goal of occupational risk assessment is often to estimate excess lifetime risk for some disabling or fatal health outcome in relation to a fixed workplace exposure lasting a working lifetime. For sub-chronic or sub-clinical health effects measured as continuous variables, the benchmark dose method can be applied, but poses issues in defining impairment and in specifying acceptable levels of excess risk. Such risks may also exhibit a dose-rate effect and partial reversibility such that effects depend on how the dose is distributed over time. Neurological

deficits as measured by a variety of increasingly sensitive neurobehavioral tests represent one such outcome, and the development of a parkinsonian syndrome among welders exposed to manganese fume presents a specific instance. Welders employed in the construction of piers for a new San Francisco-Oakland Bay Bridge in San Francisco were previously evaluated using a broad spectrum of tests. Results for four of those tests (Rey-Osterrieth Complex Figure Test, Working Memory Index, Stroop Color Word Test and Auditory Consonant Trigrams Test) were used in the benchmark dose procedure. Across the four outcomes analyzed, benchmark dose estimates were generally within a factor of 2.0, and decreased as the percentile of normal performance defining impairment increased. Estimated excess prevalence of impairment, defined as performance below the 5th percentile of normal, after 2 years of exposure at the current California standard (0.2mg/m(3), 8h TWA), ranged 15-32% for the outcomes studied. Because these exposures occurred over a 1-2-year period, generalization to lifetime excess risk requires further consideration of the form of the exposure response and whether short-term responses can be generalized to equivalent 45-year period. These results indicate unacceptable risks at the current OSHA PEL for manganese (5.0mg/m(3), 15min) and likely at the Cal OSHA PEL as well.

Kidd PM. 2005 Dec. Neurodegeneration from mitochondrial insufficiency: nutrients, stem cells, growth factors, and prospects for brain rebuilding using integrative management. Altern Med Rev 10(4):268-93.
Abstract: Degenerative brain disorders (neurodegeneration) can be frustrating for both conventional and alternative practitioners. A more comprehensive, integrative approach is urgently needed. One emerging focus for intervention is brain energetics. Specifically, mitochondrial insufficiency contributes to the etiopathology of many such disorders. Electron leakages inherent to mitochondrial energetics generate reactive oxygen free radical species that may place the ultimate limit on lifespan. Exogenous toxins, such as mercury and other environmental contaminants, exacerbate mitochondrial electron leakage, hastening their demise and that of their host cells. Studies of the brain in Alzheimer's and other dementias, Down syndrome, stroke, Parkinson's disease, multiple sclerosis, amyotrophic lateral sclerosis, Huntington's disease, Friedreich's ataxia, aging, and constitutive disorders demonstrate impairments of the mitochondrial citric acid cycle and oxidative phosphorylation (OXPHOS) enzymes. Imaging or metabolic assays frequently reveal energetic insufficiency and depleted energy reserve in brain tissue in situ. Orthomolecular nutrients involved in mitochondrial metabolism provide clinical benefit. Among these are the essential minerals and the B vitamin group; vitamins E and K; and the antioxidant and energetic cofactors alpha-lipoic acid (ALA), ubiquinone (coenzyme Q10; CoQ10), and nicotinamide adenine dinucleotide, reduced (NADH). Recent advances in the area of stem cells and growth factors encourage optimism regarding brain regeneration. The trophic nutrients acetyl L-carnitine (ALCAR), glycerophosphocholine (GPC), and phosphatidylserine (PS) provide mitochondrial support and conserve growth factor receptors; all three improved cognition in double-blind trials. The omega-3 fatty acid docosahexaenoic acid (DHA) is enzymatically combined with GPC and PS to form membrane phospholipids for nerve cell expansion. Practical recommendations are presented for integrating these safe and well-tolerated orthomolecular nutrients into a comprehensive dietary supplementation program for brain vitality and productive lifespan.

Cersosimo MG, Koller WC. 2005 Nov 30. The diagnosis of manganese-induced parkinsonism. Neurotoxicology .
Abstract: Parkinsonism is a clinical syndrome consisting of tremor, bradykinesia, rigidity, gait, balance problems, in addition to various non-motor symptoms. There are many causes of parkinsonism such as neurodegenerative disease, drugs, vascular causes, structural lesions, infections, and toxicants. Parkinson's disease, or idiopathic parkinsonism, is the most common form of parkinsonism observed in the clinic. There is degeneration of the substantia nigra, pars compacta, which results in loss

of striatal dopamine. Parkinson's disease is a slowly progressive condition in which there is a dramatic and sustained responsiveness to levodopa therapy. Manganese is an essential trace element that can be associated with neurotoxicity. Hypermanganism can occur in a variety of clinical settings. The clinical symptoms of manganese intoxication include non-specific complaints, neurobehavioral changes, parkinsonism, and dystonia. Although the globus pallidus is the main structure of damage, other basal ganglia areas can also be involved. MRI scans may show globus pallidus changes during (and for a short period after) exposure. Fluorodopa PET scans that assess the integrity of the substantia nigra dopaminergic system are abnormal in Parkinson's disease. However, these scans re-reported to be normal in a few cases studied with manganese-induced parkinsonism. The parkinsonism due to manganese may have some clinical features that occur less commonly in Parkinson's disease, such as kinetic tremor, dystonia, specific gait disturbances, and early mental, balance and speech changes. The clinical signs tend to be bilateral whereas Parkinson's disease begins on one side of the body. Patients with manganese-induced parkinsonism may be younger at the onset of the disease than with Parkinson's disease. Lastly, there appears to be a lack of response to levodopa therapy in manganese-induced parkinsonism. In summary it may be possible to differentiate manganese-induced parkinsonism from Parkinson's disease using clinical and imaging studies.

Herrero Hernandez E, Discalzi G, Valentini C, Venturi F, Chio A, Carmellino C, Rossi L, Sacchetti A, Pira E. 2005 Nov 2. Follow-up of patients affected by manganese-induced Parkinsonism after treatment with CaNa(2)EDTA. Neurotoxicology .
Abstract: In the period of 1998-2004, seven workers affected by manganese-induced Parkinsonism were diagnosed, studied and treated with CaNa(2)EDTA at our Occupational Health Ward. Biological markers, as well as magnetic resonance imaging and clinical examinations, were used to assess the disease trend. Those workers still employed were immediately removed from exposure. Our results seem to confirm that very good clinical, biological and neuroradiological results can be obtained by timely removal from exposure and chelating treatment, and that amelioration can persist in time. Manganism is, however, a severe condition that can also progress independent of further exposure. Therefore, chelating treatment can be a great aid in overt manganism, but particular attention must be paid to primary prevention, as this disease should now be totally preventable and definitely merits eradication.

Klodowska-Duda G, Jasinska-Myga B, Safranow K, Boczarska-Jedynak M, Opala G. 2005 Nov-Dec. [The role of environmental factors in Parkinson's disease may depend on disease onset age.]. Neurol Neurochir Pol 39(6):445-50.
Abstract: Background and purpose: Various factors are suspected to participate in PD onset and include environment-related factors and workplace exposure to pesticides, metals and hydrocarbons. Nevertheless, results of epidemiological research are inconsistent. Some authors emphasize hydrocarbons exposure to younger patients. Our aim was to compare PD risk factors to onset age. Material and methods: Of 174 patients with idiopathic PD, without dementia, two subgroups were isolated: 65 patients with early onset PD (EOPD) below 50 (n=65, age 52.8+/-7.6 years, onset 42.8+/-5.3 years) and 109 patients with late onset (LOPD) above 50 (n=109, age 67.8+/-7.0, onset 60.8+/-6.7 years). Various environmental factors reported in literature were analyzed. Results: The univariate analysis showed that factors significantly predisposing to EOPD are vocational education (OR 3.24, 95%CI 1.50-7.00, p<0.003), smoking (OR 1.94, 95%CI 1.02-3.69, p<0.05), well water consumption at 20-40 (OR 2.77, 95%CI 1.31-5.86, p<0.008), and after 40 (OR 4.84, 95%CI 1.95-11.99, p<0.0007), side-effects following exposure to paints (OR 2.26, 95%CI 1.10-4.66, p<0.03) and exposure to solvents (OR 1.98, 95%CI 0.96-4.07, p<0.07) on borderline significance. Drinking well water both between 20-40 and after 40 involved a substantial increase in EOPD (OR 6.57, 95%CI 2.43-17.75, p<0.0002). Education only at a primary level proved to be protective against EOPD (OR 0.20, 95%CI 0.07-0.55,

p<0.002). The multivariate logistic regression model demonstrated that independent EOPD risk factors are smoking (OR 2.20, 95%CI 1.07-4.53, p<0.04) and well water consumption both between 20-40 and after 40 (OR 8.29, 95%CI 2.73-25.23, p<0.0002), whilst the independent protective factor is education only at a primary level (OR 0.17, 95%CI 0.05-0.53, p<0.003). Conclusions: Our research demonstrated that a number of independent environmental factors significantly affect the risk of PD onset at younger ages. Presumably, some of the observed differences in the results of research of various authors into PD risk factors may be caused by ignoring onset age within the researched patients.

Kawahara M. 2005 Nov. Effects of aluminum on the nervous system and its possible link with neurodegenerative diseases. J Alzheimers Dis 8(2): 171-82; discussion 209-15.
Abstract: Aluminum is environmentally abundant, but not an essential element. Aluminum has been associated with several neurodegenerative diseases, such as dialysis encephalopathy, amyotrophic lateral sclerosis and Parkinsonism dementia in the Kii peninsula and Guam, and in particular, Alzheimer's disease. Although this association remains controversial, there is increasing evidence which suggests the implication of metal homeostasis in the pathogenesis of Alzheimer's disease. Aluminum, zinc, copper, and iron cause the conformational changes of Alzheimer's amyloid-beta protein. Al causes the accumulation of tau protein and amyloid-beta protein in experimental animals. Aluminum induces neuronal apoptosis in vivo as well as in vitro. Furthermore, a relationship between aluminum and the iron-homeostasis or calcium-homeostasis has been suggested. Based on these findings, the characteristics of aluminum neurotoxicity are reviewed, and the potential link between aluminum and neurodegenerative diseases is reconsidered.

Treiber C. 2005 Oct 26. Neurochemical insights. Sci Aging Knowledge Environ 2005(43):pe32.
Abstract: The 20th biennial meeting of the International Society for Neurochemistry was recently held in Innsbruck, Austria. This meeting gave an overview of the latest findings in the field of molecular mechanisms and diagnosis of neurodegenerative diseases such as Alzheimer's disease, Parkinson's disease, amyotrophic lateral sclerosis, and prion disease. There was a focus on the molecular pathogenesis of protein misfolding in these disorders as well as on the association between oxidative metabolism and neurological diseases. RNA interference, metal chelators, and the use of metallopeptidases were discussed as possible therapeutic strategies.

Jiang Y, Zheng W. 2005 Fall. Cardiovascular toxicities upon manganese exposure. Cardiovasc Toxicol 5(4):345-54.
Abstract: Manganese (Mn)-induced Parkinsonism has been well documented; however, little attention has been devoted to Mn-induced cardiovascular dysfunction. This review summarizes literature data from both animal and human studies on Mn's effect on cardiovascular function. Clinical and epidemiological evidence suggests that the incidence of abnormal electrocardiogram (ECG) is significantly higher in Mn-exposed workers than that in the control subjects. The main types of abnormal ECG include sinus tachycardia, sinus bradycardia, sinus arrhythmia, sinister megacardia, and ST-T changes. The accelerated heart-beat and shortened P-R interval appear to be more prominent in female exposed workers than in their male counterparts. Mn-exposed workers display a mean diastolic blood pressure that is significantly lower than that of the control subjects, especially in the young and female exposed workers. Animal studies indicate that Mn is capable of quickly accumulating in heart tissue, resulting in acute or subacute cardiovascular disorders, such as acute cardiodepression and hypotension. These toxic outcomes appear to be associated with Mn-induced mitochondrial damage and interaction with the calcium channel in the cardiovascular system.

Aschner JL, Aschner M. 2005 Aug-Oct. Nutritional aspects of manganese

homeostasis. Mol Aspects Med 26(4-5):353-62.
Abstract: Manganese (Mn) is an essential mineral. It is present in virtually all diets at low concentrations. The principal route of intake for Mn is via food consumption, but in occupational cohorts, inhalation exposure may also occur (this subject will not be dealt with in this review). Humans maintain stable tissue levels of Mn. This is achieved via tight homeostatic control of both absorption and excretion. Nevertheless, it is well established that exposure to high oral, parenteral or ambient air concentrations of Mn can result in elevations in tissue Mn levels. Excessive Mn accumulation in the central nervous system (CNS) is an established clinical entity, referred to as manganism. It resembles idiopathic Parkinson's disease (IPD) in its clinical features, resulting in adverse neurological effects both in laboratory animals and humans. This review focuses on an area that to date has received little consideration, namely the potential exposure of parenterally fed neonates to exceedingly high Mn concentrations in parenteral nutrition solutions, potentially increasing their risk for Mn-induced adverse health sequelae. The review will consider (1) the essentiality of Mn; (2) the concentration ranges, means and variation of Mn in various foods and infant formulas; (3) the absorption, distribution, and elimination of Mn after oral exposure and (4) the factors that raise a theoretical concern that neonates receiving total parenteral nutrition (TPN) are exposed to excessive dietary Mn.

Weinreb O, Amit T, Bar-Am O, Chillag-Talmor O, Youdim MB. 2005 Aug. Novel Neuroprotective Mechanism of Action of Rasagiline Is Associated with Its Propargyl Moiety: Interaction of Bcl-2 Family Members with PKC Pathway. Ann N Y Acad Sci 1053:348-55.
Abstract: Our studies have provided new insights into the biological mechanism of neuroprotection of the anti-Parkinson drug, rasagiline [N-propargyl-(1R)-aminoindan], involving the association of Bcl-2 family proteins with protein kinase C (PKC) pathway. In a model of serum withdrawal-induced apoptosis of rat pheochromocytoma PC12 cells, rasagiline and its propargyl moiety, N-propargylamine, decreased cell death via multiple neuroprotective pathways that include the stimulation of PKC phosphorylation; upregulation of PKCepsilon mRNA; induction of Bcl-X (L), Bcl-w, and brain-derived neurotrophic factor (BDNF) mRNAs; and downregulation of PKCgamma, Bad, and Bax mRNAs. Moreover, these drugs inhibited the cleavage and activation of pro-caspase-3 and poly(ADP-ribose) polymerase (PARP), while PKC inhibitor, GF109203X, reversed these actions. In addition, rasagiline decreased serum-free-induced levels of the important regulator of cell death, Bad, which was also blocked by GF109203X, indicating the involvement of PKC-dependent cell survival activity of rasagiline. Structure activity studies have established that N-propargylamine is essential for the novel neuroprotective and the neuronal cell survival activity of rasagiline since this moiety itself revealed similar protective effects and mechanisms of action. These results have led us to develop several multifunctional neuroprotective drugs containing the propargyl moiety and iron-chelating property for the treatment and/or prevention of neurodegenerative diseases.

Trenkwalder C, Paulus W, Walters AS. 2005 Aug. The restless legs syndrome. Lancet Neurol 4(8):465-75.
Abstract: The restless legs syndrome is a common disorder that encompasses an idiopathic form of genetic or unknown origin and symptomatic forms associated with many causes. Symptomatic forms occur during pregnancy and are coincident with uraemia, iron depletion, polyneuropathy, spinal disorders, and rheumatoid arthritis. For the hereditary forms, at least three gene loci, located on chromosomes 12, 14, and 9, have been traced so far. Prevalence in the general population is between 3% and 9%, increases with age, and is higher in women than in men. Treatment is needed only in the moderate to severe forms of the disorder and mostly in elderly people. Pathophysiology and treatment may be closely linked to the dopaminergic system and iron metabolism. Dopaminergic treatment with levodopa and dopamine agonists is the first choice in idiopathic restless legs syndrome, but augmentation and rebound

should be monitored in long-term treatment. Various other drugs, such as opioids, gabapentin, and benzodiazepines, provide alternative treatment possibilities.

Mandel S, Grunblatt E, Riederer P, Amariglio N, Jacob-Hirsch J, Rechavi G, Youdim MB. 2005 Aug. Gene Expression Profiling of Sporadic Parkinson's Disease Substantia Nigra Pars Compacta Reveals Impairment of Ubiquitin-Proteasome Subunits, SKP1A, Aldehyde Dehydrogenase, and Chaperone HSC-70. Ann N Y Acad Sci 1053:356-75 .
Abstract: Sporadic Parkinson's disease (PD) constitutes 99% of the disorder, while the remaining 1% of the cases is of familial (genetic) origin. The mutations reported to be associated with familial PD indicate impairment in protein processing and misfolding, as is handled by the ubiquitin-proteasome system (UPS), and in mitochondrial function. For these reasons, we have recently applied, for the first time, Affymetrix oligonucleotide microarray technique in the substantia nigra pars compacta of sporadic parkinsonian patients for studying global gene expression analysis and comparison to the alterations identified in inherited PD. This study identified decreased expression of 68 genes and elevation of 69 genes. Classification into functional groups revealed that the downregulated genes are related to signal transduction, protein degradation (e.g., ubiquitin-proteasome subunits), dopaminergic transmission/metabolism, iron transport, protein modification/phosphorylation, and energy pathways/glycolysis functional classes. A major finding is the decreased expressions of 5 subunits of the UPS, SKP1A, a member of the SCF (E3) ubiquitin ligase complex, and chaperone HSC-70, which can lead to a wide impairment in the function of an entire repertoire of proteins. The upregulated genes are clustered in cell adhesion/cytoskeleton, extracellular matrix components, cell cycle, protein modification/phosphorylation, protein metabolism and transcription, and inflammation/hypoxia (e.g., key iron and oxygen sensor EGLN1) classes. The study shows, for the first time, a convergence in the pathogenic processes that are observed in hereditary (familial) and sporadic PD, where abnormal iron metabolism, oxidative stress, and aggregation of proteins occur. An additional breakthrough in this research is the identification of a number of previously unsuspected crucial gene players that are also involved in the process of neurodegeneration, which can serve as specific biomarkers for PD and novel drug development.

Kosta P, Argyropoulou MI, Markoula S, Konitsiotis S. 2005 Jun 29. MRI evaluation of the basal ganglia size and iron content in patients with Parkinson's disease. J Neurol .
Abstract: OBJECTIVE: To evaluate by MRI the area size and the degree of iron accumulation in basal ganglia nuclei that are implicated in the pathogenesis of Parkinson's disease (PD).METHODS: 40 patients with idiopathic PD and 40 controls were examined on a 1. 5 Tesla MR imager, using a multiecho SE sequence 2000/20, 40, 60, 80, 100, 120, 140, 160 (TR/TE). The T2 relaxation time (T2) and the area of substantia nigra zona compacta (SNc), substantia nigra zona reticulata (SNr), putamen (Pu), globus pallidus external (GPe), globus pallidus internal (GPi), caudate nucleus (CN), locus coeruleus (LC) and subthalamic nucleus (STN) were assessed.RESULTS: The T2 of SNc (76. 8 +/- 6. 0) was lower and of Pu (79.5 +/- 6.0) and GPe (69.5 +/- 7.0) was higher in patients than in controls (78.6 +/- 3.8, 77.4 +/- 3.9 and 67.3 +/- 5.7, respectively), $p < 0.05$. The area of CN (125.9 +/- 20.2) and Pu (201.5 +/- 48.7) was higher in patients than in controls (110.7 +/- 21.5 and 180.1 +/- 41.1, respectively), $p < 0.05$. A more pronounced decrease in the T2 of SNc (73.6 +/- 8.9) was observed when the more affected side of patients was evaluated separately. In patients with disease duration > 5 years the T2 of STN (71.5 +/- 6.3) was lower and the area of Pu was higher (215.3 +/- 54.9) compared with those with disease duration </= 5 years (75.8 +/- 10.9 and 190.9 +/- 41.0 respectively), $p < 0.05$.CONCLUSIONS: These findings suggest that dysfunction of the basal ganglia circuitry in PD may affect iron content not only in SNc but in STN, Pu and GPe as well. Compensatory sprouting of the remaining dopaminergic fibers could

account for the increased area of the CN and Pu.

Weiss B. 2005 Jun 1. Economic Implications of Manganese Neurotoxicity. Neurotoxicology .
Abstract: Manganese neurotoxicity is linked primarily to inhalation exposure, and its clinical features are almost totally based on high doses, such as those experienced by miners. Manifestations of lower level exposures can take two forms. One is the appearance of neurobehavioral deficits. A second, equally subtle, form is as a promoter, borrowing the term used in carcinogenesis, of neurodegenerative disease. Such low-level environmental exposures may be more potent than expected if they occur as ultrafine particles able to penetrate directly into the brain. The neurological disorder linked most closely to manganese is Parkinson's disease (PD). Although most observers recognize that the features of manganese-induced parkinsonism differ from those of idiopathic PD, they overlap considerably. The overlaps should be expected because the underlying lesions, although distinguishable, are closely linked because they belong to structures with complex interdependent circuitry. Such interdependence makes it feasible to undertake an analysis of how manganese neurotoxicity might elevate the risks of PD. A relatively small increment in risk, expressed as a leftward shift in the age prevalence of PD, incurs significant economic costs.

Shinbo Y, Taira T, Niki T, Iguchi-Ariga SM, Ariga H. 2005 Mar. DJ-1 restores p53 transcription activity inhibited by Topors/p53BP3. Int J Oncol 26(3):641-8.
Abstract: DJ-1 is a multi-functional protein that plays roles in transcriptional regulation and anti-oxidative stress, and loss of its function is thought to result in onset of Parkinson's disease. Here, we report that DJ-1 bound to Topors/p53BP3, a ring finger protein binding to both topoisomerase I and p53, in vitro and in vivo and that both proteins were colocalized in cells. DJ-1 and p53 were then found to be sumoylated by Topors in cells. It was also found that DJ-1 bound to p53 in vitro and in vivo and that colocalization with and its binding to p53 were stimulated by UV irradiation of cells. Transcription activity of p53 was found to be abrogated by Topors concomitant with sumoylation of p53 in a dose-dependent manner, and DJ-1 restored its repressed activity by releasing the sumoylated form of p53. These findings suggest that DJ-1 positively regulates p53 through Topors-mediated sumoylation.

Zhou Y, Wang Y, Kovacs M, Jin JH, Zhang J. 2005. Microglial activation induced by neurodegeneration - A proteomic analysis. Molecular & Cellular Proteomics 4(10):1471-1479.
Abstract: Neuroinflammation mediated by microglial activation appears to play an essential role in the pathogenesis of Parkinson disease; however, the mechanisms by which microglia are activated are not fully understood. Thus, we first evaluated the effects of two parkinsonian toxicants, manganese ethylene bisdithiocarbamate (Mn-EBDC) and 1-methyl-4-phenylpyridine (MPP+), on microglial activation as well as associated dopaminergic (DAergic) neurotoxicity in primary cell culture systems. The results demonstrated that, when rat primary mesencephalic neuron-enriched or neuron-microglia mixed cultures were treated with Mn-EBDC at 2 - 8 mu M or MPP+ at 0.25 - 5 mu M, respectively, for 7 days, both toxicants were capable of inducing DAergic neurodegeneration as well as activating microglia via a mechanism secondary to DAergic neurodegeneration. Furthermore activated microglia subsequently enhanced DAergic neurotoxicity induced by Mn- EBDC or MPP+. Detailed scrutiny of neuron-microglia interactions identified a fraction of the conditioned media derived from a DAergic cell line treated with Mn- EBDC or MPP+ that potently activated microglia. To further define potential mediators leading to microglial activation secondary to neurodegeneration, we utilized a quantitative proteomic technique termed SILAC (for stable isotope labeling by amino acids in cell culture) to compare the protein profiles of MPP+-treated cellular fraction that mediated microglial activation as compared with controls. The search revealed numerous novel proteins that are potentially important in neurodegeneration-mediated microglial

activation, a process believed to be critical in Parkinson disease progression.

Zheng HL, Youdim MBH, Weiner LM, Fridkin M. 2005. Novel potential neuroprotective agents with both iron chelating and amino acid-based derivatives targeting central nervous system neurons. Biochem Pharmacol 70(11):1642-1652.
Abstract: Antioxidants and iron chelating molecules are known as neuroprotective agents in animal models of neurodegenerative disorders such as Alzheimer's disease (AD) and Parkinson's disease (PD). In this study, we designed and synthesized a novel bifunctional molecule (M10) with radical scavenging and iron chelating ability on an amino acid carrier likely to be a substrate for system L, thus targeting the compound to the central nervous system (CNS). M10 had a moderate iron affinity in HEPES buffer (pH 7.4) with log K-3 = 12.25 +/- 0.55 but exhibited highly inhibitory action against iron-induced lipid peroxidation, with an IC50 value (12 mu M) comparable to that of desferal (DFO). EPR studies indicated that M10 was a highly potent (center dot)0H scavenger with an IC50 of about 0.3 molar ratio of M10 to H2O2. In PC12 cell culture, M10 was at least as potent as the anti-Parkinson drug rasagiline in protecting against cell death induced by serum-deprivation and by 6-hydroxydopamine (6-OHDA). These results suggest that M10 deserves further investigation as a potential agent for the treatment of neurodegenerative disorders such as AD and PD. (c) 2005 Elsevier Inc. All rights reserved.

Zheng HL, Weiner LM, Bar-Am O, Epsztejn S, Cabantchik ZI, Warshawsky A, Youdim NBH, Fridkin M. 2005. Design, synthesis, and evaluation of novel bifunctional iron-chelators as potential agents for neuroprotection in Alzheimer's, Parkinson's, and other neurodegenerative diseases. Bioorganic & Medicinal Chemistry 13(3):773-783.
Abstract: Several novel antioxidant-iron chelators bearing 8-hydroxyoxyquinoline moiety were synthesized, and various properties related to their iron chelation, and neuroprotective action were investigated. All the chelators exhibited strong iron(III) chelating and high antioxidant properties. Chelator 9 (HLA20). having good permeability into K562 cells and moderate selective MAO-B inhibitory activity (IC50 110 muM), displayed the hightest protective effects against differentiated P19 cell death induced by 6-hydroxydopamine. EPR studies suggested that Chelator 9 also act as radical scavenger to directly scavenge hydroxyl radical. (C) 2004 Elsevier Ltd. All rights reserved.

Zheng H, Youdim MBH, Weiner LM, Fridkin M. 2005. Synthesis and evaluation of peptidic metal chelators for neuroprotection in neurodegenerative diseases. J Pept Res 66(4):190-203.
Abstract: A series of novel derivatives of neuropeptides with a metal-chelating moiety was synthesized and examined for various properties related to iron (Fe) chelation and neuroprotective action. All derivatives chelated Fe to form stable Fe complexes in water. Some strongly inhibited Fe-induced lipid peroxidation with an IC50 value of about 12 mu M. In PC12 cell culture, several compounds, at concentrations as low as 1 mu M, attenuated serum-free stimulated cell death and improved cell survival by 20-35%. At this concentration, these analogs also protected against 6-hydroxydopamine (6-OHDA)-induced cell death, increasing cell viability by 20-30%. Electron paramagnetic resonance (EPR) studies indicated that besides being good Fe chelators, these analogs act as radical scavengers to directly scavenge hydroxyl radicals. Together, the data indicate that some of the analogs could be further developed as possible neuroprotective agents for treatment of neurodegenerative diseases such as Parkinson's, Alzheimer's, and Huntington's diseases, Friedreich's atxia, amyotrophic, and lateral sclerosis where Fe misregulation has been reported.

Zheng H, Gal S, Weiner LM, Bar-Am O, Warshawsky A, Fridkin M, Youdim MBH. 2005. Novel multifunctional neuroprotective iron chelator-monoamine oxidase inhibitor drugs for neurodegenerative diseases: in vitro studies on

antioxidant activity, prevention of lipid peroxide formation and monoamine oxidase inhibition. J Neurochem 95(1):68-78.
Abstract: Iron-dependent oxidative stress, elevated levels of iron and of monoamine oxidase (MAO)-B activity, and depletion of antioxidants in the brain may be major pathogenic factors in Parkinson's disease, Alzheimer's disease and related neurodegenerative diseases. Accordingly, iron chelators, antioxidants and MAO-B inhibitors have shown efficacy in a variety of cellular and animal models of CNS injury. In searching for novel antioxidant iron chelators with potential MAO-B inhibitory activity, a series of new iron chelators has been designed, synthesized and investigated. In this study, the novel chelators were further examined for their activity as antioxidants, MAO-B inhibitors and neuroprotective agents in vitro. Three of the selected chelators (M30, HLA20 and M32) were the most effective in inhibiting iron-dependent lipid peroxidation in rat brain homogenates with IC50 values (12-16 mu M), which is comparable with that of desferal, a prototype iron chelator that is not has orally active. Their antioxidant activities were further confirmed using electron paramagnetic resonance spectroscopy. In PC12 cell culture, the three novel chelators at 0.1 mu M were able to attenuate cell death induced by serum deprivation and by 6-hydroxydopamine. M30 possessing propargyl, the MAO inhibitory moiety of the anti-Parkinson drug rasagiline, displayed greater neuroprotective potency than that of rasagiline. In addition, in vitro, M30 was a highly potent non-selective MAO-A and MAO-B inhibitor (IC50 < 0.1 mu M). However, HLA20 was more selective for MAO-B but had poor MAO inhibition, with an IC50 value of 64.2 mu M. The data suggest that M30 and HLA20 might serve as leads in developing drugs with multifunctional activities for the treatment of various neurodegenerative disorders.

Zhang XS, Haaf M, Todorich B, Grosstephan E, Schieremberg H, Surguladze N, Connor JR. 2005. Cytokine toxicity to oligodendrocyte precursors is mediated by iron. Glia 52(3):199-208.
Abstract: Inflammatory processes play a key role in the pathogenesis of a number of common neurodegenerative disorders such as Alzheimer's disease (AD), Parkinson's disease (PD), and multiple sclerosis (MS). Abnormal iron accumulation is frequently noted in these diseases and compelling evidence exists that iron is involved in inflammatory reactions. Histochemical stains for iron repeatedly demonstrate that oligodendrocytes, under normal conditions, stain more prominently than any other cell type in the brain. Therefore, we examined the hypothesis that cytokine toxicity to oligodendrocytes is iron mediated. Oligodendrocytes in culture were exposed to interferon-gamma (IFN-gamma), interleukin-1 beta (IL-1 beta), and tumor necrosis factor-alpha (TNF-alpha). Toxicity was observed in a dose-dependent manner for IFN-gamma and TNF-alpha. IL-1 beta was not toxic in the concentrations used in this study. The toxic concentration of IFN-gamma, and TNF-alpha was lower if the cells were iron loaded, but iron loading had no effect on the toxicity of IL-1 beta. These data provide insight into the controversy regarding the toxicity of cytokines to oligodendrocytes by revealing that iron status of these cells will significantly impact the outcome of cytokine treatment. The exposure of oligodendrocytes to cytokines plus iron decreased mitochondrial membrane potential but activation of caspase 3 is limited. The antioxidant, TPPB, which targets mitochondria, protected the oligodendrocytes from the iron-mediated cytotoxicity, providing further support that mitochondrial dysfunction may underlie the iron-mediated cytokine toxicity. Therapeutic strategies involving anti-inflammatory agents have met with limited success in the treatment of demyelinating disorders. A better understanding of these agents and the contribution of cellular iron status to cytokine toxicity may help develop a more consistent intervention strategy. (c) 2005 Wiley-Liss, Inc.

Zhang X, Xie WJ, Qu S, Pan TH, Wang XT, Le WD. 2005. Neuroprotection by iron chelator against proteasome inhibitor-induced nigral degeneration. Biochem Biophys Res Commun 333(2):544-549.
Abstract: The cause of the neurodegenerative process in Parkinson's disease (PD) remains unclear, but evidence suggests that failure of the

ubiquitin-proteasome system may play a major role in the pathogenesis of the disease. Iron is believed to be a key contributor to PD pathology by inducing aggregation of a-synuclein and by generating oxidative stress. Our present studies have shown that micro-injection of the proteasome inhibitor lactacystin into the substantia nigra (SN) of C57BL/6 mice causes significant loss of dopaminergic cells and induces intracellular inclusion body formation. We have also found that co-injection of the iron chelator desferrioxamine not only attenuates the lactacystin-induced dopamine neuron loss, but also reduces the presence of ubiquitin-positive intracellular inclusions in the SN, whereas use of iron-deficient diet has no such protective effects. These results may support that iron plays a key role in proteasome inhibitor-induced nigral pathology and that reducing iron reactivity may prevent dopaminergic neuron degeneration and reduce abnormal protein aggregation. (c) 2005 Elsevier Inc. All rights reserved.

Zhang PJ, Land W, Lee S, Juliani J, Lefman J, Smith SR, Germain D, Kessel M, Leapman R, Rouault TA, Subramaniam S. 2005. Electron tomography of degenerating neurons in mice with abnormal regulation of iron metabolism. J Struct Biol 150(2):144-153.
Abstract: Previous studies have shown that IRP1(+/-) IRP2(-/-) knockout mice develop progressive neurodegenerative symptoms similar to those observed in human movement disorders such as Parkinson's disease. Histological investigations using optical microscopy show that these IRP knockout mice display accumulation of ferritin in axonal tracts in the brain, suggesting a possible role for excess ferritin in mediating axonal degeneration. Direct observation of the 3D distribution of ferritin by electron tomography indicates that ferritin amounts are increased by 3- to 4-fold in selected regions of the brain, and structural damage is observed within the axon as evidenced by the loss of the internal network of filaments, and the invaginations of neighboring oligodendrocyte membranes into the axonal medium. While optical microscopic investigations suggest that there is a large increase in ferritin in the presumptive axonal regions of the IRP knockout mice, electron tomographic studies reveal that most of the excess ferritin is localized to double-walled vesicular compartments which are present in the interior of the axon and appear to represent invaginations of the oligodendrocyte cells into the axon. The amount of ferritin observed in the axonal space of the knockout mice is at least 10-fold less than the amount of ferritin observed in wild-type mouse axons. The surprising conclusion from our analysis, therefore, is that despite the overall increase in ferritin levels in the knockout mouse brain, ferritin is absent from axons of degenerating neurons, suggesting that trafficking is compromised in early stages of this type of neuronal degeneration. Published by Elsevier Inc.

Zhang J, Stanton DM, Nguyen XV, Liu M, Zhang Z, Gash D, Bing G. 2005. Intrapallidal lipopolysaccharide injection increases iron and ferritin levels in glia of the rat substantia nigra and induces locomotor deficits. Neuroscience 135(3):829-838.
Abstract: Increasing evidence suggests that abnormal iron handling may be involved in the pathogenesis of Parkinson's disease. The present study investigates the role of iron and the iron-storage protein ferritin in inflammation-induced degeneration of dopaminergic neurons of the substantia nigra pars compacta. Injection of lipopolysaccharide into the globus pallidus of young and middle-aged rats substantially decreased tyrosine hydroxylase immunostaining in substantia nigra pars compacta four weeks after injection. Loss of tyrosine hydroxylase expression was accompanied by increased iron and ferritin levels in glial cells of the substantia nigra pars reticulata. Despite greater increases in nigral iron levels, ferritin induction was less pronounced in older rats, suggesting the regulation of ferritin was compromised with age. Automated movement tracking analyses showed that young rats recovered from LPS-induced locomotor deficits within four weeks, yet older rats failed to improve on measures of speed and total distance moved. Intrapallidal lipopolysaccharide injection also increased expression of alpha-synuclein and ubiquitin in tyrosine hydroxylase-positive neurons of the substantia

nigra pars compacta. These results suggest that pallidal inflammation significantly increases stress on dopamine-containing neurons in the substantia nigra pars compacta. Alterations in nigral iron levels and protein handing may increase the vulnerability of nigral neurons to degenerative processes. (c) 2005 Published by Elsevier Ltd on behalf of IBRO.

Zecca L, Zucca FA, Toscani M, Adorni F, Giaveri G, Rizzio E, Gallorini M. 2005. Iron, copper and their proteins in substantia nigra of human brain during aging. Journal of Radioanalytical and Nuclear Chemistry 263(3):733-737.
Abstract: Scarce information is available on the content of metals and their molecules in the human brain. Iron, copper and other metals are involved in neurodegenerative disorders like Parkinson' s Disease (PD), however, their behavior in physiological conditions is poorly understood. In this study we have measured iron, copper and their major proteins (ferritins and ceruloplasmin) in substantia nigra (SN) of normal subjects at different ages, since this is the main target region of PD. An increasing trend for iron and copper concentration was found in aging. Ferritins were also increasing in aging while ceruloplasmin did not vary. These data show that the accumulation of these metals requires an increased expression of storage molecules to prevent toxic effects of iron and copper.

Zecca L, Berg D, Arzberger T, Ruprecht P, Rausch WD, Musicco M, Tampellini D, Riederer P, Gerlach M, Becker G. 2005. In vivo detection of iron and neuromelanin by transcranial sonography: A new of substantia approach for early detection nigra damage. Mov Disord 20(10):1278-1285.
Abstract: Early diagnosis of Parkinson's disease (PD) in nonsymptomatic patients is a key issue. An increased echogenicity of the substantia nigra (SN) was found previously in Parkinsonian patients and in a low percentage of healthy adults. These nonsymptomatic subjects also showed a reduced F-18-dopa uptake in striatum, suggesting a preclinical injury of the nigrostriatal system that could later proceed into PD. To investigate the ability of ultrasonography to detect markers of SN degeneration, such as iron deposition and neuromelanin depletion, we scanned postmortem brains from normal subjects at different ages by ultrasound and measured the echogenic area of the SN. The SN was then dissected and used for histological examinations and determination of iron, ferritin, and neuromelanin content. A significant positive correlation was found between the echogenic area of the SN and the concentration of iron, H- and L-ferritins. Multivariate analysis carried out considering the iron content showed a significant negative correlation between echogenicity and neuromelanin content of the SN. In PD, a typical loss of neuromelanin and increase of iron is observed in this brain area. The finding of a positive correlation between iron and ferritin levels and a negative correlation of neuromelanin content with the area of echogenicity at the SN could therefore provide an interesting basis for diagnosis and therapeutic follow-up studies in PD. (c) 2005 Movement Disorder Society.

Zahir F, Rizwi SJ, Haq SK, Khan RH. 2005. Low dose mercury toxicity and human health. Environ Toxicol Pharmacol 20(2):351-360.
Abstract: Post Minamata incident there has been awareness about mercury toxicity even among the general public. Previous researches contributed a vast amount of data regarding acute mercury exposure, but gradually information about the low dose [Ninomiya, T., Ohmori, H., Hashimoto, K., Tsuruta, K., Ekino, S., 1995. Expansion of methylmercury poisoning outside minamata: an epidemiological study on chronic methylmercury poisoninig outside of Minamata. Environ. Res. 70 (1) 47-50; Lebel, J., Mergler, D., Lucotte, M., Amorim, M., Dolbec, J., Miranda, D., Arantes, G., Rheault, L, Pichet, P., 1996. Evidence of early nervous system dysfunction in Amazonian populations exposed to low-levels of methylmercury. Neurotoxicology 17 (1) 157-167] of mercury toxicity has been trickling in. With mercury contaminating rain-, ground and sea-water no one is safe. Polluted water leads to mercury laced fish, meat and vegetable. In aquatic environments, inorganic mercury is microbiologically transformed into lipophilic organic compound 'methylmercury'. This transformation makes mercury more prone to biomagnification in food chains. Consequently,

populations with traditionally high dietary intake of food originating from fresh or marine environment have highest dietary exposure to mercury. Extensive research done on locals across the globe have already established this, persons who routinely consume fish or a particular species of fish are at an increased risk of methylmercury poisoning. The easy access of the toxicant to man through multiple pathways air, water, food, cosmetic products and even vaccines increase the exposure. Foetus and children are more susceptible towards mercury toxicity. Mothers consuming diet containing mercury pass the toxicant to foetus and to infants through breast milk. Decreased performance in areas of motor function and memory has been reported among children exposed to presumably safe mercury levels. Similarly, disruption of attention, fine motor function and verbal memory was also found in adults on exposure to low mercury levels. It is an occupational hazard for dental staff, chloralkali factory workers and goldminers, etc. Mercury has been found to be a causative agent of various sorts of disorders, including neurological, nephrological, immunological, cardiac, motor, reproductive and even genetic. Recently heavy metal mediated toxicity has been linked to diseases like Alzeihemer's, Parkinson's, Autism, Lupus, Amyotrophic lateral sclerosis, etc. Besides this, it poses danger to wildlife. Therefore, it becomes imperative to spread the information regarding the threat of mercury exposure amongst the scientists and masses.

Youdim MBH, Fridkin M, Zheng HL. 2005. Bifunctional drug derivatives of MAO-B inhibitor rasagiline and iron chelator VK-28 as a more effective approach to treatment of brain ageing and ageing neurodegenerative diseases. Mech Ageing Dev 126(2):317-326.
Abstract: Degeneration of nigrostriatal dopamine neurons and cholinergic cortical neurones are the main pathological features of Parkinson's disease (PD) and for the cognitive deficit in dementia of the Alzheimer' type (AD) and in dementia with Lewy bodies (DLB), respectively. Many PD and DLB subjects have dementia and depression resulting from possible degeneration of cholinergic and noradrenergic and serotonergic neurons. On the other hand, AD patients may also develop extrapyramidal features as well as depression. In both PD and AD there is, respectively, accumulation of iron within the melanin containing dopamine neurons of pars compacta and with in the plaques and tangle. It has been suggested that iron accumulation may contribute to the oxidative stress induced apoptosis reported in both diseases. This may result from increased glia hydrogen peroxide producing monoamine oxidase (MAO) activity that can generate of reactive hydroxyl radical formed from interaction of iron and hydrogen peroxide. We have therefore prepared a series of novel bifunctional drugs from the neuroprotective-antiapoptotic antiparkinson monoamine oxidase B inhibitor, rasagiline, by introducing a carbamate cholinesterase (ChE) inhibitory moiety into it. Ladostigil (TV-3326, N-propargyl-3R-aminoindan-5yl)-ethyI methylcarbamate), has both ChE and MAO-AB inhibitory activity, as potential treatment of AD and DLB or PD subjects with dementia Being a brain selective MAO-AB inhibitor it has limited potentiation of the pressor response to oral tyramine and exhibits antidepressant activity similar to classical non-selective MAO inhibitor antidepressants by increasing brain serotonin and noradrenaline. Ladostigil inhibits brain acetyl and butyrylcholinesterase in rats and antagonizes scopolamine-induced inhibition of spatial learning. Ladostigil like MAO-B inhibitor it prevents MPTP Parkinsonism in mice model and retains the in vitro and in vivo neuroprotective activity of rasagiline. Ladostigil, rasagiline and other propargylamines have been demonstrated to have neuroprotective activity in several in vitro and in vivo models, which have been shown be associated with propargylamines moiety, since propargylamines itself possess these properties. The mechanism of neuroprotective activity has been attributed to the ability of propargylarnines-inducing the antiapoptotic family proteins Bcl-2 and Bcl-xl, while decreasing Bad and Bax and preventing opening of mitochondrial permeability transition pore. Iron accumulates in brain regions associated with neurodegenerative diseases of PD, AD, amyotrophic lateral sclerosis and Huntington disease. It is thought to be involved in Fenton chemistry

oxidative stress observed in these diseases. The neuroprotective activity of propargylamines led us to develop several novel bifunctional iron chelator from our prototype brain permeable iron chelators, VK-28, possessing propargylamine moiety (HLA-20, M30 and M30A) to iron out iron from the brain. These compounds have been shown to have iron chelating and monoamine oxidase A and B selective brain inhibitory and neuroprotective-antiapoptotic actions. (C) 2004 Elsevier Ireland Ltd. All rights reserved.

Yavin E, Kikkiri R, Gil S, Arad-Yellin R, Yavin E, Shanzer A. 2005. Synthesis and biological evaluation of lipophilic iron chelators as protective agents from oxidative stress. Organic & Biomolecular Chemistry 3(15):2685-2687.
Abstract: Lipophilic Fe-III chelators were synthesized and shown to protect oligodendrial cells from oxidative damage induced by Fe-III and hydrogen peroxide.

Yamin G, Munishkina LA, Karymov MA, Lyubchenko YL, Uversky VN, Fink AL. 2005. Forcing nonamyloidogenic beta-synuclein to fibrillate. Biochemistry (Mosc) 44(25):9096-9107.
Abstract: The fibrillation and aggregation of alpha-synuclein is a key process in the formation of intracellular inclusions, Lewy bodies, in substantia nigral neurons and, potentially, in the pathology of Parkinson's disease and several other neurodegenerative disorders. alpha-Synuclein and its homologue P-synuclein are both natively unfolded proteins that colocalize in presynaptic terminals of neurons in many regions of the brain, including those of dopamine-producing cells of the substantia nigra. Unlike its homologue, P-synuclein does not form fibrils and has been shown to inhibit the fibrillation of alpha-synuclein. In this study, we demonstrate that fast and efficient aggregation and fibrillation of beta-synuclein can be induced in the presence of a variety of factors. Certain metals (Zn2+, Pb2+, and Cu2+) induce a partially folded conformation of beta-synuclein that triggers rapid fibrillation. In the presence of these metals, mixtures of alpha- and beta-synucleins exhibited rapid fibrillation. The metal-induced fibrillation of beta-synuclein was further accelerated by the addition of glycosaminoglycans or high concentrations of macromolecular crowding agents. beta-Synuclein also rapidly formed soluble oligomers and fibrils in the presence of pesticides, whereas the addition of low concentrations of organic solvents induced formation of amorphous aggregates. These new findings demonstrate the potential effect of environmental pollutants in generating an amyloidogenic, and potentially neurotoxic, conformation, in an otherwise benign protein.

Yamamoto A, Friedlein A, Imai Y, Takahashi R, Kahle PJ, Haass C. 2005. Parkin phosphorylation and modulation of its E3 ubiquitin ligase activity. J Biol Chem 280(5):3390-3399.
Abstract: Mutations in the PARKIN gene are the most common cause of hereditary parkinsonism. The parkin protein comprises an N-terminal ubiquitin-like domain, a linker region containing caspase cleavage sites, a unique domain in the central portion, and a special zinc finger configuration termed RING-IBR-RING. Parkin has E3 ubiquitin-protein ligase activity and is believed to mediate proteasomal degradation of aggregation-prone proteins. Whereas the effects of mutations on the structure and function of parkin have been intensely studied, post-translational modifications of parkin and the regulation of its enzymatic activity are poorly understood. Here we report that parkin is phosphorylated both in human embryonic kidney HEK293 cells and human neuroblastoma SH-SY5Y cells. The turnover of parkin phosphorylation was rapid, because inhibition of phosphatases with okadaic acid was necessary to stabilize phosphoparkin. Phosphoamino acid analysis revealed that phosphorylation occurred mainly on serine residues under these conditions. At least five phosphorylation sites were identified, including Ser(101), Ser(131), and Ser(136) (located in the linker region) as well as Ser(296) and Ser(378) (located in the RING-IBR-RING motif). Casein kinase-1, protein kinase X and protein kinase C phosphorylated parkin in vitro, and inhibition of casein kinase-1 caused a dramatic reduction of parkin phosphorylation in cell lysates. Induction of protein folding stress in cells reduced parkin phosphorylation,

and unphosphorylated parkin had slightly but significantly elevated autoubiquitination activity. Thus, complex regulation of the phosphorylation state of parkin may contribute to the unfolded protein response in stressed cells.

Xiang ZX, Wang L, Kitai ST. 2005. Modulation of spontaneous firing in rat subthalamic neurons by 5-HT receptor subtypes. J Neurophysiol 93(3): 1145-1157.
Abstract: The subtlialamic nucleus (STN) is considered to be one of the driving forces in the basal ganglia circuit. The STN is innervated by serotoneraic afferents from the raphe nucleus and expresses a variety of 5-HT receptor subtypes. We investigated the effects of 5-HT and 5-HT receptor subtype agonists and antagonists on the firing properties of STN neurons in rat brain slices. We used cell-attached, perforated-patch. and whole cell recording techniques to detect changes in firms! frequency and pattern and electrical membrane properties. Due to the depolarization of membrane potential caused by reduced potassium conductance, 5-HT (10 muM) increased. the firing frequency of STN neurons without changing their firing pattern. Cadmium failed to occlude the effect of 5-HT on firing frequency. 5-HT had no effect on afterhyperpolarization current. These results indicated that the 5-HT action was not mediated by high-voltage-activated calcium channel currents and calcium-dependent potassium currents. 5-HT had no effect on hyperpolarization-activated cation current (I-H) amplitude and voltage-dependence of I,, activation, suggesting that I was not involved in 5-HT-induced excitation. The H increased firing by 5-HT was mimicked by 5-HT,, receptor agonist alpha-methyl-5-HT and was partially mimicked by 5-HT2 receptor agonist DOI or 5-HT4 receptor agonist cisapride. The 5-HT action was partially reversed by 5-HT4 receptor antagonist SB 23597-190, 5-HT2 receptor antagonist ketanserin, and 5-HT2, receptor antagonist RS 102221. Our data indicate that 5-HT has significant ability to modulate membrane excitability in STN neurons; modulation is accomplished by decreasing potassium conductance by activating 5-HT4 and 5-HT2c receptors.

Xiang MH, Mohamalawari D, Rao R. 2005. A novel isoform of the secretory pathway Ca2+, Mn2+-ATPase, hSPCA2, has unusual properties and is expressed in the brain. J Biol Chem 280(12):11608-11614.
Abstract: Unlike lower eukaryotes, mammalian genomes have a second gene, ATP2C2, encoding a putative member of ;the family of secretory pathway Ca2+, Mn2+-ATPases, SPCA2. Human SPCA2 shares 64% amino acid identity with the protein defective in Hailey Hailey disease, hSPCA1. We show that human SPCA2 (hSPCA2) has a more limited tissue distribution than hSPCA1, with prominent protein expression in brain and testis. In primary neuronal cells, endogenous SPCA2 has a highly punctate distribution that overlaps with vesicles derived from the trans-Golgi network and is thus different from the compact perinuclear distribution of hSPCA1 seen in keratinocytes and nonpolarized cells. Heterologous expression in a yeast strain lacking endogenous Ca2+ pumps reveals further functional differences from hSPCA1. Although the Mn2+-specific phenotype of hSPCA2 is similar to that of hSPCA1, Ca2+ ions are transported with much poorer affinity, resulting in only weak complementation of Ca2+-specific yeast phenotypes. These observations suggest that SPCA2 may have a more specialized role in mammalian cells, possibly in cellular detoxification of Mn2+ ions, similar to that in yeast. We point to the close links between manganese neurotoxicity and Parkinsonism that would predict an important physiological role for SPCA2 in the brain.

Wielgus AR, Sarna T. 2005. Melanin in human irides of different color and age of donors. Pigment Cell Res 18(6):454-464.
Abstract: Melanin is the main chromophore of the human iris. This pigment is considered to be the most important factor that determines the color of the irides. Previous studies based mainly on chemical degradation methods showed that brown irides contain more melanin than blue ones. In our study, we used electron spin resonance (ESR) spectroscopy to detect and

characterize melanin free radical centers and associated iron in human irides. Based on this method, we determined the amount of melanin in the irides and the relative content of iron in iridial melanin as a function of their color, shade, and the age of their donors. Chemical degradation of iridial homogenates enabled us to characterize the structure of eumelanin and determine the content of pheomelanin present in human and bovine irides. The ESR amplitude, the normalized intensity obtained by double integration of the ESR signal of melanin, and the content of the pigment in the irides depended on color and shade of the eyes being 40% higher in the brown group of the irides compared with all other groups. On the other hand, the relative iron content normalized to the melanin content in light blue irides showed a small decrease with age of donors. Melanin in human and bovine irides was mostly composed of eumelanin, and pheomelanin content was of the order of a few percent. Although some differences in the structure of eumelanin present in the human and bovine irides are possible, the results obtained in this study suggest that human irides contain eumelanin with very similar chemical properties.

Wang C, Ko HS, Thomas B, Tsang F, Chew KCM, Tay SP, Ho MWL, Lim TM, Soong TW, Pletnikova O, Troncoso J, Dawson VL, Dawson TM, Lim KL. 2005. Stress-induced alterations in parkin solubility promote parkin aggregation and compromise parkin's protective function. Hum Mol Genet 14(24): 3885-3897.
Abstract: Mutations in parkin are currently recognized as the most common cause of familial Parkinsonism. Emerging evidence also suggests that parkin expression variability may confer a risk for the development of the more common, sporadic form of Parkinson's disease (PD). Supporting this, we have recently demonstrated that parkin solubility in the human brain becomes altered with age. As parkin apparently functions as a broad-spectrum neuroprotectant, the resulting decrease in the availability of soluble parkin with age may underlie the progressive susceptibility of the brain to stress. Interestingly, we also observed that many familial-PD mutations of parkin alter its solubility in a manner that is highly reminiscent of our observations with the aged brain. The converging effects on parkin brought about by aging and PD-causing mutations are probably not trivial and suggest that environmental modulators affecting parkin solubility would increase an individual's risk of developing PD. Using both cell culture and in vivo models, we demonstrate here that several PD-linked stressors, including neurotoxins (MPP+, rotenone, 6-hydroxydopamine), paraquat, NO, dopamine and iron, induce alterations in parkin solubility and result in its intracellular aggregation. Furthermore, the depletion of soluble, functional forms of parkin is associated with reduced proteasomal activities and increased cell death. Our results suggest that exogenously introduced stress as well as endogenous dopamine could affect the native structure of parkin, promote its misfolding, and concomitantly compromise its protective functions. Mechanistically, our results provide a link between the influence of environmental and intrinsic factors and genetic susceptibilities in PD pathogenesis.

Walter U, Krolikowski K, Tarnacka B, Benecke R, Czlonkowska A, Dressler D. 2005. Sonographic detection of basal ganglia lesions in asymptomatic and symptomatic Wilson disease. Neurology 64(10):1726-1732.
Abstract: Objective: To investigate whether transcranial brain parenchyma sonography (TCS) detects basal ganglia abnormalities in asymptomatic and symptomatic patients with Wilson disease (WD) and whether findings correlate with disease severity. Methods: Twenty-one patients with WD with (n = 18) or without (n = 3) neurologic symptoms were investigated. Disease severity was assessed by three independent neurologists using a WD rating scale (WDRS) with the items dysarthria, akinesia, ataxia, tremor, and dystonia; the raters' median score was used for further analysis. Basal ganglia TCS was performed according to a standardized protocol. Results: TCS revealed lenticular nucleus (LN) hyperechogenicity in all assessable neurologically symptomatic and in two of the three asymptomatic patients. Size of LN hyperechogenic area correlated with the WDRS score (Spearman correlation, rho = 0.604, p = 0.006), as did the

size of thalamus hyperechogenic area (n = 7, rho = 0.891, p = 0.007), the width of third ventricle (n = 21, rho = 0.613, p = 0.003), and the width of lateral ventricles (n = 20, rho = 0.642, p < 0.001). Substantia nigra hyperechogenicity, detected in 10 patients, did not correlate with disease severity. There was no correlation between age at disease onset or disease duration and any TCS finding. Of the 19 patients with LN hyperechogenicity, only 12 showed abnormal LN on MRI. Conclusions: Transcranial brain parenchyma sonography (TCS) detects lenticular nucleus hyperechogenicity, likely to be caused by copper accumulation, in neurologically symptomatic and asymptomatic Wilson disease (WD). TCS findings correlate with disease severity. TCS appears a promising tool for disease monitoring in WD.

Vestergaard M, Kerman K, Tamiya E. 2005. An electrochemical approach for detecting copper-chelating properties of flavonoids using disposable pencil graphite electrodes: Possible implications in copper-mediated illnesses. Anal Chim Acta 538(1-2):273-281.
Abstract: We have studied the electrochemistry of eight flavonoids belonging to four flavonoid sub-classes: flavone, flavonol, flavanol and anthocyanidin using pencil graphite electrodes (PGEs). We present the electrochemistry of delphinidin, cyanidin and catechin gallate for the first time. The use of electrochemical methods in connection with PGE in the study of flavonoids and their interaction with copper ions has not been previously reported. Our results compare favorably with previously reported studies, which utilised glassy carbon electrodes (GCEs) for the detection of flavonoids. We calibrated all eight flavonoids (r(2) > 0.9620), six of them at at least two peak potentials. The relative standard deviation (R.S.D.) for peak potential was < 5.0% and peak height was < 10.0%; thus, this method could be used to characterise and quantify flavonoid-containing extracts (purified). An inverse relationship between oxidation potential and metal-chelation was established. Oxidation potential was influenced by the location of OH groups relative to each other, the oxidation state of the pyranose ring, the presence of a C-4-oxo group and the total number of OH groups. Further, we showed that the steric configuration of the compound influenced the reactivity. The order of flavonoid reactivity to Cu(II) ions was myricertin = catechin gallate > quercetin > delphinidin = baicalein > cyanidin > catechin. These findings may be significant in neuroscience and metal toxicological studies, in which copper ions have been reported to play a crucial role in initiating and/or promoting the progression of diseases such as Alzheimer's and Parkinson's. © 2005 Elsevier B.V. All fights reserved.

Ved R, Saha S, Westlund B, Perier C, Burnam L, Sluder A, Hoener M, Rodrigues CMP, Alfonso A, Steer C, Liu L, Przedborski S, Wolozin B. 2005. Similar patterns of mitochondrial vulnerability and rescue induced by genetic modification of alpha-synuclein, parkin, and DJ-1 in Caenorhabditis elegans. J Biol Chem 280(52):42655-42668.
Abstract: How genetic and environmental factors interact in Parkinson disease is poorly understood. We have now compared the patterns of vulnerability and rescue of Caenorhabditis elegans with genetic modifications of three different genetic factors implicated in Parkinson disease (PD). We observed that expressing alpha-synuclein, deleting parkin (K08E3.7), or knocking down DJ-1 (B0432.2) or parkin produces similar patterns of pharmacological vulnerability and rescue. C. elegans lines with these genetic changes were more vulnerable than nontransgenic nematodes to mitochondrial complex I inhibitors, including rotenone, fenperoximate, pyridaben, or stigmatellin. In contrast, the genetic manipulations did not increase sensitivity to paraquat, sodium azide, divalent metal ions (Fe(II) or Cu(II)), or etoposide compared with the nontransgenic nematodes. Each of the PD-related lines was also partially rescued by the antioxidant probucol, the mitochondrial complex II activator, D-beta-hydroxybutyrate, or the anti-apoptotic bile acid tauroursodeoxycholic acid. Complete protection in all lines was achieved by combining D-beta-hydroxybutyrate with tauroursodeoxycholic acid but not with probucol. These results show that diverse PD-related genetic

modifications disrupt the mitochondrial function in C. elegans, and they raise the possibility that mitochondrial disruption is a pathway shared in common by many types of familial PD.

Vassiliev V, Harris ZL, Zatta P. 2005. Ceruloplasmin in neurodegenerative diseases. Brain Research Reviews 49(3):633-640.
Abstract: For decades, abnormalities in ceruloplasmin (Cp) synthesis have been associated with neurodegenerative disease. From the early observation that low circulating serum ceruloplasmin levels served as a marker for Wilson's disease to the recent characterization of a neurodegenerative disorder associated with a complete lack of serum ceruloplasmin, the link between Cp and neuropathology has strengthened. The mechanisms associated with these different central nervous system abnormalities are very distinct. In Wilson's disease, a defect in the P-type ATPase results in abnormal hepatic copper accumulation that eventually leaks into the circulation and is abnormally deposited in the brain. In this case, copper deposition results in the neurodegenerative phenotype observed. Patients with autosomal recessive condition, aceruloplasminemia, lack the ferroxidase activity inherent to the multi-copper oxidase ceruloplasmin and develop abnormal iron accumulation within the central nervous system. In the following review ceruloplasmin gene expression, structure and function will be presented and the role of ceruloplasmin in iron metabolism will be discussed. The molecular events underlying the different forms of neurodegeneration observed will be presented. Understanding the role of ceruloplasmin within the central nervous system is fundamental to further our understanding of the pathology observed. Is the ferroxidase function more essential than the antioxidant role? Does Cp help maintain nitrosothiol stores or does it oxidize critical brain substrates? The answers to these questions hold the promise for the treatment of devastating neurodegenerative conditions such as Alzheimer's and Parkinson's diseases. It is essential to further elucidate the mechanism of the neuronal injury associated with these disorders. (c) 2005 Elsevier B.V. All rights reserved.

Vanlandingham JW, Tassabehji NM, Somers RC, Levenson CW. 2005. Expression profiling of p53-target genes in copper-mediated neuronal apoptosis. Neuromolecular Medicine 7(4):311-324.
Abstract: Copper toxicity associated with Wilson's disease is known to cause neuronal damage and death in the basal ganglia and frontal cortex leading to Parkinson-like symptoms and cognitive deficits. Our previous work in cultured human NTERA-2-N neurons showed that copper-induced neuronal apoptosis is dependent on the induction and nuclear translocation of the tumor suppressor protein, p53. Because p53 acts as a DNA-binding transcription factor, this work used an oligonucleotide array to identify p53 target genes that are differentially regulated in copper-loaded neurons. Arrays representing 145 human genes expressed downstream of p53 were hybridized with labeled mRNA from control and copper-treated neurons. Differentially regulated mRNAs included those involved in the regulation of the cell cycle, cytoprotective mechanisms, and apoptotic mechanisms. Transfection of cells with a dominant-negative p53 construct enabled us to determine which molecular events were dependent on p53 expression. Copper treatment resulted in the upregulation of p21, reprimo, stathmin, and Tp531NP1, all known to participate in cell cycle arrest. Protective mechanisms included the upregulation of stat-3, and the heat-shock proteins, heat-shock protein (Hsp) 70 and Hsp 27. Both p53-dependent and -independent mechanisms leading to apoptosis were identified including insulin-like growth factor binding protein-6, glutathione peroxidase, bcl-2, RB-1, PUMA, and several members of the redox active PIG family of proteins. Thus it appears that following copper-mediated neuronal DNA damage, the regulation of a variety of pro- and antiapoptotic genes are responsible for determining neuronal fate.

Uversky VN, Yamin G, Munishkina LA, Karymov MA, Millett IS, Doniach S, Lyubchenko YL, Fink AL. 2005. Effects of nitration on the structure and aggregation of alpha-synuclein. Molecular Brain Research 134(1):84-102.

Abstract: Substantial evidence suggests that the aggregation of the presynaptic protein alpha-synuclein is a key step in the etiology of Parkinson's disease (PD). Although the molecular mechanisms underlying alpha-synuclein aggregation remain unknown, oxidative stress has been implicated in the pathogenesis of PD. Here, we report the effects of tyrosine nitration on the propensity of human recombinant a-synuclein to fibrillate in vitro. The properties of nitrated alpha-synuclein were investigated using a variety of biophysical and biochemical techniques, which revealed that nitration led to formation of a partially folded conformation with increased secondary structure relative to the intrinsically disordered structure of the monomer, and to oligomerization at neutral pH. The degree of self-association was concentration-dependent, but at 1 mg/mL, nitrated alpha-synuclein was predominantly an octamer. At low pH, small-angle X-ray scattering data indicated that the nitrated protein was monomeric. alpha-Synuclein fibrillation at neutral pH was completely inhibited by nitrotyrosination and is attributed to the formation of stable soluble oligomers. The presence of heparin or metals did not overcome the inhibition; however, the inhibitory effect was eliminated at low pH. The addition of nitrated alpha-synuclein inhibited fibrillation of non-modified alpha-synuclein at neutral pH. Potential implications of these findings to the etiology of Parkinson's disease are discussed. (C) 2004 Elsevier B.V All rights reserved.

Tjoa CW, Benedict RHB, Weinstock-Guttman B, Fabiano AJ, Bakshi R. 2005. MRI T2 hypointensity of the dentate nucleus is related to ambulatory impairment in multiple sclerosis. J Neurol Sci 234(1-2):17-24.
Abstract: Objectives: MRI T2 hypointensity in multiple sclerosis (MS) gray matter, suggesting iron deposition, is associated with physical disability, disease course, lesion load, and brain atrophy. Ambulatory dysfunction limits quality of life; however correlation with conventional MRI remains poor. Methods: Normalized intensity on T2-weighted images was obtained in the basal ganglia, thalamus, red nucleus, and dentate nucleus in 47 MS patients and 15 healthy controls. Brain T1-hypointense and FLAIR-hyperintense lesion volume, third ventricle width, brain parenchymal fraction and timed 25 foot walk (T25FW) were measured in the MS group. Results: T2 hypointensity was present throughout gray matter in MS vs. controls (all $p < 0.01$). Dentate T2 hypointensity was the only MRI variable significantly correlated with T25FW (Pearson $r=-0.355$, $p=0.007$) and was also the best MRI correlate of physical disability (EDSS) score in regression modeling ($r=-0.463$, $R-2=0.223$, $p=0.004$). Conclusions: T2 hypointensity is present in subcortical gray matter nuclei in patients with MS vs. normal controls. Dentate nucleus T2 hypointensity is independently related to ambulatory impairment and disability, accounting for more variance than conventional lesion and atrophy measures. This study adds more weight to the notion that T2 hypointensity is a clinically relevant marker of tissue damage in MS. (c) 2005 Elsevier B.V. All rights reserved.

Thiel S, Heinson G, White A. 2005. Tectonic evolution of the southern Gawler Craton, South Australia, from electromagnetic sounding. Australian Journal of Earth Sciences 52(6):887-896.
Abstract: Long-period natural-source electromagnetic data have been recorded using portable three-component magnetometers at 39 sites in 1998 and 2002 across the southern Eyre Peninsula, South Australia that forms part of the Gawler Craton. Site spacing was of order 5 km, but reduced to I km or less near known geological boundaries, with a total survey length of approximately 50 km. A profile trending east-west was inverted for a 2D electrical resistivity model to a depth of 20 km across the southern Eyre Peninsula. The main features from the models are: (1) on the eastern side of the Gawler Craton, the Donington Suite granitoids to the east of the Kalinjala Shear Zone are resistive (>1000 Omega m); (ii) the boundary between the Donington Suite granitods and the Archaean Sleaford Complex, which has much lower resistivity of 10-100 Omega m, is almost vertical in the top 10 km and dips slightly westwards; and (III) two very low resistivity (< 1 Omega m) arcuate zones in the top 3 km of Hutchison Group sediments correlate with banded iron-formations, and are

probably related to biogenic-origin graphite deposits concentrated in fold hinges, Such features suggest an extensional regime during the time period 2.00-1.85 Ga. We suggest that the resistivity boundary between the Donington Suite and the Archaean Sleaford Complex represents a growth fault, typical for rift systems that evolve into a half-graben structure. In the graben basin, low-resistivity shallow-marine Hutchison Group sediments were deposited. Folding of the sediments during the Kimban Orogeny between 1.74 and 1.70 Ga has led to migration of graphite to the fold hinges resulting in linear zones of very low resistivity that correlate with banded iron-formation magnetic anomalies.

Tharakan B, Dhanasekaran M, Manyam BV. 2005. Antioxidant and DNA protecting properties of anti-fatigue herb Trichopus zeylanicus. Phytother Res 19(8): 669-673.
Abstract: Chronic fatigue is considered a complex symptom for which currently there is no curative treatment available. Oxidative stress plays an important role in the etiology of fatigue and antioxidant treatment might be a valuable therapeutic approach. The Kani, a tribal high altitude living population in southern India, traditionally use the seeds of Trichopus zeylanicus to combat fatigue. In this study, the antioxidant properties of Trichopus zeylanicus were established on free radicals (DPPH and ABTS), its ability to reduce iron, lipoxygenase activity and hydrogen peroxide-induced lipid peroxidation. The effects of Trichopus zeylanicus on reactive oxygen species induced plasmid DNA (pBR322) cleavage were also investigated. Trichopus zeylanicus significantly scavenged free radicals, reduced lipid peroxidation and inhibited lipoxygenase activity. Trichopus zeylanicus also exhibited iron-chelating activity and inhibited reactive oxygen species induced DNA damage. Trichopus zeylanicus contains NADH, polyphenols and sulfhydryl compounds, which have the ability to scavenge reactive oxygen species suggesting that the antioxidant activity may be an important mechanism of action of Trichopus zeylanicus to combat fatigue. Copyright (c) 2005 John Wiley & Sons, Ltd.

Tarohda T, Ishida Y, Kawai K, Yamamoto M, Amano R. 2005. Regional distributions of manganese, iron, copper, and zinc in the brains of 6-hydroxydopamine-induced parkinsonian rats. Analytical and Bioanalytical Chemistry 383(2):224-234.
Abstract: Time courses of changes in manganese, iron, copper, and zinc concentrations were examined in regions of the brain of a 6-hydroxydopamine (6-OHDA)-induced rat model of Parkinson's disease using inductively coupled plasma mass spectrometry (ICP-MS). The concentrations were simultaneously determined in brain section at the level of the substantia nigra 1, 3, 7, 10, 14 and 21 days after the 6-OHDA treatment and compared with those of control rats. The distributions of these elements were obtained for 18 regions of the sagittal section (1-mm thick). The ICP-MS results indicated that Mn, Fe, Cu, and Zn levels of the 6-OHDA-induced parkinsonian brain were observed to increase in all regions that lay along the dopaminergic pathway. In the substantia nigra, the increase in Mn level occurred rapidly from 3 to 7 days and preceded those in the other elements, reaching a plateau in the 6-OHDA brain. Iron and Zn levels increased gradually until 7 days and then increased rapidly from 7 to 10 days. The increase in the

Tabner BJ, El-Agnaf OMA, German MJ, Fullwood NJ, Allsop D. 2005. Protein aggregation, metals and oxidative stress in neurodegenerative diseases. Biochem Soc Trans 33:1082-1086.
Abstract: There is clear evidence implicating oxidative stress in the pathology of many different neurodegenerative diseases. ROS (reactive oxygen species) are the primary mediators of oxidative stress and many of the aggregating proteins and peptides associated with neurodegenerative disease can generate hydrogen peroxide, a key ROS, apparently through interactions with redox-active metal ions. our recent results suggest that ROS are generated during the very early stages of protein aggregation, when protofibrils or soluble oligomers are present, but in the absence of mature amyloid fibrils. The generation of ROS during

early-stage protein aggregation may be a common, fundamental molecular mechanism underlying the pathogenesis of oxidative damage, neurodegeneration and cell death in several different neurodegenerative diseases. Drugs that specifically target this process could be useful in the future therapy of these diseases.

Szczerbowska-Boruchowska M, Chwiej J, Lankosz M, Adamek D, Wojcik S, Krygowska-Wajs A, Tomik B, Bohic S, Susini J, Simionovici A, Dumas P, Kastyak M. 2005. Intraneuronal investigations of organic components and trace elements with the use of synchrotron radiation. X-Ray Spectrometry 34(6):514-520.
Abstract: Three techniques based on synchrotron radiation microbeam analysis were applied to biochemical investigations of human central nervous system (CNS) tissue. Thin tissue slices representing Parkinson's disease (PD), amyotrophic lateral sclerosis (ALS) and control cases were studied. Synchrotron radiation microbeam x-ray fluorescence (micro-SRXRF) was applied to the analysis of the elemental distribution inside single nerve cells. Investigation of copper oxidation state was performed with the use of micro x-ray absorption near-edge structure spectroscopy (micro-XANES). Moreover, the organic components of the tissues were analyzed by means of synchrotron radiation Fourier transform infrared microspectroscopy (SR-FTIRM). Topographic differences in elemental composition were observed for the nerve cell body. Higher levels of Fe, Zn and Ca were found for the neurons of PD cases in comparison with the control group. The IR spectra measured in neurons of PD, ALS and control cases showed differences in absorption bands associated with characteristic groups of selected biological molecules. Additionally, results of SR-FTIRM measurements indicated inhomogeneous accumulation of the main organic components in PD nerve cells, in contrast to the control cases. Copyright (c) 2005 John Wiley & Sons, Ltd.

Sutoh Y, Nishida Y. 2005. Formation of a Mn(IV) species in the reaction mixture of a manganese(II) complex and an aliphatic aldehyde. Synthesis and Reactivity in Inorganic Metal-Organic and Nano-Metal Chemistry 35(7): 575-577.
Abstract: ESR spectra showed that manganese(II) complexes with tripodal-ligands containing a benzimidazole group can activate oxygen easily in the presence of an aliphatic aldehyde leading to the conversion of the Mn(II) species to a Mn(IV) species. The above facts may give important information to elucidate the origin of the so-called "manganism," a dreaded illness marked by Parkinson's-like tremors, and also give possible reasons as to why the soil in certain regions of Iceland, Slovakia, and Colorado where scrapie, Creutzfeldt-Jacob disease, and chronic wasting disease clusters are found, is low in copper and high in manganese.

Sung JY, Park SM, Lee CH, Um JW, Lee HJ, Kim J, Oh YJ, Lee ST, Paik SR, Chung KC. 2005 . Proteolytic cleavage of extracellular secreted alpha-synuclein via matrix metalloproteinases. J Biol Chem 280(26):25216-25224.
Abstract: Although alpha-synuclein is the main structural component of the insoluble filaments that form Lewy bodies in Parkinson disease (PD), its physiological function and exact role in neuronal death remain poorly understood. In the present study, we examined the possible functional relationship between alpha-synuclein and several forms of matrix metalloproteinases (MMPs) in the human dopaminergic neuroblastoma (SK-N-BE) cell line. When SK-N-BE cells were transiently transfected with alpha-synuclein, it was secreted into the extracellular culture media, concomitantly with a significant decrease in cell viability. Also the addition of nitric oxide-generating compounds to the cells caused the secreted alpha-synuclein to be digested, producing a small fragment whose size was similar to that of the fragment generated during the incubation of alpha-synuclein with various MMPs in vitro. Among several forms of MMPs, alpha-synuclein was cleaved most efficiently by MMP-3, and MALDI-TOF mass spectra analysis showed that alpha-synuclein is cleaved from its C-terminal end with at least four cleavage sites within the non-A beta component of AD amyloid sequence. Compared with the intact form, the protein

aggregation of alpha-synuclein was remarkably facilitated in the presence of the proteolytic fragments, and the fragment-induced aggregates showed more toxic effect on cell viability. Moreover, the levels of MMP-3 were also found to be increased significantly in the rat PD brain model produced by the cerebral injection of 6-hydroxydopamine into the substantia nigra. The present study suggests that the extracellularly secreted alpha-synuclein could be processed via the activation of MMP-3 in a selective manner.

Stroh A, Faber C, Neuberger T, Lorenz P, Sieland K, Jakob PM, Webb A, Pilgrimm H, Schober R, Pohl EE, Zimmer C. 2005. In vivo detection limits of magnetically labeled embryonic stem cells in the rat brain using high-field (17.6 T) magnetic resonance imaging. Neuroimage 24(3):635-645.
Abstract: Stem cell transplantation is a promising therapeutic approach for several neurological disorders. However, it has yet to fulfill its high expectations, partially due to the lack of a reliable noninvasive method for monitoring the biodistribution of the grafted stem cells in vivo. We have used high-resolution magnetic resonance imaging (MRI) at 17.6 T, combined with efficient magnetic labeling of the stem cells with iron oxide nanoparticles. in order to assess the in vivo detection limit in small animal models. Injection of different concentrations of magnetically labeled stem cells in gel phantoms led to significant reductions in image intensity from small cellular clusters of less than 10 cells. To determine the detection limit in vivo, various numbers of both labeled and unlabeled cells were injected stereotactically into the striatum of rats. Significant hypointense signal changes were observed for 100 labeled cells. After injection of approximately 20 labeled cells, signal reduction at the injection site was observed but could not be assigned unambiguously to the cells. Our results show that high-field MRI allows tracking of a minimal number of cells in vivo, well below the number used in previous studies. opening the possibility of gaining new insights into cell migration and differentiation. (C) 2004 Elsevier Inc. All rights reserved.

Spencer PS, Palmer VS, Ludolph AC. 2005. On the decline and etiology of high-incidence motor system disease in West Papua (southwest New Guinea). Mov Disord 20:S119-S126.
Abstract: The etiology of a high-incidence focus of amyotrophic lateral sclerosis and parkinsonism-dementia (ALS/P-D) in south West Papua (Irian Jaya, Indonesia), first described in the 1960s and 1970s, has been attributed to mineral deficiencies, hyperparathyroidism, and metal neurotoxicity arising from reliance on drinking water obtained from springs and shallow wells. More recent visits (1987 and 1990) to the south West Papua focus of neurodegenerative disease cast doubt on this explanation by revealing changes in disease prevalence in communities with an unchanged water supply. These communities have experienced a dramatic decline in ALS and a reversal in the relative prevalence of ALS and parkinsonism. The extrapyramidal disorder can be distinguished from Parkinson disease by pyramidal features (and dementia) reminiscent of Guam P-D. Topical use of cycad seed (termed kurru) gametophyte to treat large skin lesions is advanced as a plausible but unproven etiologic factor. Medicinal use of untreated cycad seed (Cycas sp.) has also been linked with ALS foci in Japan (oral use) and Guam (topical use), with the additional consumption on Guam of food items prepared from Cycas sp. seed or animals that consume cycad seed components. (c) 2005 Movement Disorder Society.

Smith RR, Dimayuga ER, Keller JN, Maragos WF . 2005. Enhanced toxicity to the catecholamine tyramine in polyglutamine transfected SH-SY5Y cells. Neurochem Res 30(4):527-531.
Abstract: Huntington's disease (HD) is a progressive neurodegenerative disorder, of which the pathogenesis is not completely understood. In patients with Huntington's disease, there is a mutation in the gene encoding the protein huntingtin, which results in an expanded polyglutamine sequence leading to degeneration of the basal ganglia. There is mounting evidence that metabolism of the transmitter dopamine by the enzyme monoamine oxidase may contribute to striatal damage in mitochondrial

toxin-induced models of HD. In this study, we have examined the role of the catecholamine tyramine in neural SH-SY5Y cells transfected with normal and expanded polyglutamine repeat numbers. Our findings demonstrate that cells containing a pathological number of polyglutamines are more sensitive to tyramine than cells with a non-pathological number. Tyramine-induced cell death was attenuated by MAO inhibitors as well as with catalase and the iron chelator deferoxamine, suggesting that H2O2 might mediate the observed toxicity. These observations support the notion that the metabolism of dopamine plays a role in neuron death in Huntington's disease.

Simmons CR, Hao Q, Stipanuk MH. 2005. Preparation, crystallization and X-ray diffraction analysis to 1.5 angstrom resolution of rat cysteine dioxygenase, a mononuclear iron enzyme responsible for cysteine thiol oxidation. Acta Crystallographica Section F-Structural Biology and Crystallization Communications 61:1013-1016.

Siddiq A, Ayoub IA, Chavez JC, Aminova L, Shah S, Lamanna JC, Patton SM, Connor JR, Cherny RA, Volitakis I, Bush AI, Langsetmo I, Seeley T, Gunzler V, Ratan RR. 2005. Hypoxia-inducible factor prolyl 4-hydroxylase inhibition - A target for neuroprotection in the central nervous system. J Biol Chem 280(50):41732-41743.
Abstract: Hypoxia-inducible factor (HIF) prolyl 4-hydroxylases are a family of iron- and 2-oxoglutarate-dependent dioxygenases that negatively regulate the stability of several proteins that have established roles in adaptation to hypoxic or oxidative stress. These proteins include the transcriptional activators HIF-1 alpha and HIF-2 alpha. The ability of the inhibitors of HIF prolyl 4-hydroxylases to stabilize proteins involved in adaptation in neurons and to prevent neuronal injury remains unclear. We reported that structurally diverse low molecular weight or peptide inhibitors of the HIF prolyl 4-hydroxylases stabilize HIF-1 alpha and up-regulate HIF-dependent target genes (e.g. enolase, p21(waf1/cip1), vascular endothelial growth factor, or erythropoietin) in embryonic cortical neurons in vitro or in adult rat brains in vivo. We also showed that structurally diverse HIF prolyl 4-hydroxylase inhibitors prevent oxidative death in vitro and ischemic injury in vivo. Taken together these findings identified low molecular weight and peptide HIF prolyl 4-hydroxylase inhibitors as novel neurological therapeutics for stroke as well as other diseases associated with oxidative stress.

Shackelford RE, Manuszak RP, Heard SC, Link CJ, Wang SM. 2005. Pharmacological manipulation of ataxia-telangiectasia kinase activity as a treatment for Parkinson's disease. Med Hypotheses 64(4):736-741.
Abstract: Parkinson's disease (PD) is a major cause of morbidity and mortality among older individuals. Although the causes of Parkinson's disease are multifactorial, considerable evidence indicates that elevated labile iron in the substantia nigra pars compacta plays an important role in producing oxyradicals which subsequently damage nigro-striatat neurons. Based on this several researchers have suggested that blood-brain barrier crossing iron chelators might have clinical efficacy in treating PD. Work demonstrating that iron chelators protect nigro-striatat neurons in the N-methyL-4-phenyl-1,2,3,6-tetrahydropyridine and 6-hydroxydopamine-induced rodent PD models supports this hypothesis. Recently, we found that the ATM gene product (mutated in ataxia-telangiectasia, A-T), is required for cell survival and genomic stability maintenance following exposure to low labile iron concentrations. Iron chelators (desferal, quercetin, and apoferritin) also increase A-T cell. genomic stability and viability, and activate ATM-dependent cellular events in normal cells. Additionally Atm-deficient mice exhibit a selective loss of dopaminergic nigro-striatal. neurons. Based on this, we propose that iron chelators protect the substantia nigra pars compacta not only by chelating labile iron and reducing oxyradical formation, but also by inducing ATM activity, leading to increased oxidative stress resistance and DNA repair. Support for this hypothesis comes from the recent observation that the iron chelating flavonoid quercetin both directly activates ATM and protects

neuronal cells from the toxic effects of the N-methyl-4-phenyl-1,2,3,6-tetrahydropyridine. Therefore since; (1) ATM is required for iron toxicity resistance, (2) iron chelators such as quercetin, desferal, and apoferritin induce ATM activity and/or ATM-dependent events, and (3), Atm-deficient mice preferentially lose dopaminergic nigro-striatat neurons, we propose that ATM activity has an important function in PD. Furthermore, pharmacological. manipulation of ATM activity via iron chelation might have clinical efficacy in PD treatment. (c) 2004 Elsevier Ltd. All rights reserved.

Schroder JM. 2005. Ferritinopathy: diagnosis by muscle or nerve biopsy, with a note on other nuclear inclusion body diseases. Acta Neuropathol (Berl) 109(1):109-114.
Abstract: Ferritinopathy (neuroferritinopathy) has recently been identified as an autosomal dominant, multisystem disease, mainly affecting the central nervous system. It is caused by mutations in exon 4 of the ferritin light chain gene on chromosome 19. Its fine structural hallmarks are granular nuclear inclusions in neurons, oligodendroglial and microglial cells with similar extracellular derivatives in the central nervous system, muscle, peripheral nerve, and skin. These pathognostic structures have previously been described in perivascular cells of muscle and nerve biopsy specimens in a case with an obviously identical disease, formerly described as 'granular nuclear inclusion body disease'. The nuclear inclusions, at the light microscopic level, are iron positive following histochemical iron reactions and immunoreactive for ferritin antibodies. At the electron microscopic level, in contrast to filamentous nuclear inclusions in 'neuronal intranuclear hyaline inclusion disease', dominant spinocerebellar atrophies and other trinucleotide repeat diseases, they are basically composed of granules measuring 5-15 nm. A moderate peak of iron detectable by energy dispersive microanalysis of the granular nuclear inclusions in ferritinopathy may also be significant. It is emphasized that ferritinopathy or 'granular nuclear inclusion body disease' can be diagnosed by a simple muscle or nerve biopsy without brain biopsy, autopsy, or molecular genetic testing of the considerable number of neurodegenerative diseases with possibly similar symptomatology.

Schmidauer C, Sojer M, Seppi M, Stockner H, Hogl B, Biedermann B, Brandauer E, Peralta CM, Wenning GK, Poewe W. 2005. Transcranial ultrasound shows nigral hypoechogenicity in restless legs syndrome. Ann Neurol 58(4): 630-634.
Abstract: In patients with Parkinson's disease, hyperechogenicity of the substantia nigra using transcranial ultrasound has been related to increased tissue concentrations of iron. Recently, deficient iron transport mechanisms in substantia nigra neurons have been described in postmortem tissue of patients with restless legs syndrome (RLS). This study was performed to study substantia nigra echogenicity in RLS patients compared with normal control subjects and Parkinson's disease patients. RLS patients had significantly reduced midbrain areas of hyperechogenicity compared with control subjects, and even more markedly reduced hyperechogenicity compared with Parkinson's disease patients. These findings lend further support to nigral iron deficiency as a pathogenetic factor in RLS.

Sayre LM, Moreira PI, Smith MA, Perry G. 2005. Metal ions and oxidative protein modification in neurological disease. Ann Ist Super Sanita 41(2):143-164.
Abstract: This review highlights the role of oxidative stress and imbalances in metal ion homeostasis in the neurodegenerative diseases Alzheimer's disease and Parkinson's disease and in the progressive demyelinating disease multiple sclerosis. The chemistry and biochemistry of oxidative stress-induced protein damage are first described, followed by the evidence for a pathological role of oxidative stress in these disease states. It is tempting to speculate that free radical oxygen chemistry contributes to pathogenesis in all these conditions, though it is as yet undetermined what types of oxidative changes occur early in the disease, and what types are secondary manifestations of neuronal degeneration.

Sanchez P, Galvez N, Colacio E, Minones E, Dominguez-Vera JM. 2005. Catechol releases iron(III) from ferritin by direct chelation without iron(II) production. Dalton Transactions (4):811-813.
Abstract: It has been traditionally considered that catechols release iron from ferritin by reduction to iron(II), which diffuses through the ferritin channels into the intracellular milieu where it participates in the Fenton reaction, producing highly toxic hydroxyl radicals. However, in the present work we have proved that the mechanism of the release of iron from ferritin by catechol does not take place by iron(II) reduction but by direct iron(III) chelation and therefore without iron(II) production. A possible extension of these findings to other catechols is discussed on the basis of the stability with respect to the internal redox reaction of the iron(III) catechol complexes.

Samuele A, Mangiagalli A, Armentero MT, Fancellu R, Bazzini E, Vairetti M, Ferrigno A, Richelmi P, Nappi G, Blandini F. 2005. Oxidative stress and pro-apoptotic conditions in a rodent model of Wilson's disease. Biochimica Et Biophysica Acta-Molecular Basis of Disease 1741(3):325-330.
Abstract: Wilson's disease (WD) is an inherited disorder, characterized by selective copper deposition in liver and brain, chronic hepatitis and extrapyramidal signs. In this study, we investigated changes of biochemical markers of oxidative stress and apoptosis in liver, striatum and cerebral cortex homogenates from Long-Evans Cinnamon (LEC) rats, a mutant strain isolated from Long Evans (LE) rats, in whom spontaneous hepatitis develops shortly after birth. LEC and control (LE) rats at I I and 14 weeks of age were used. We determined tissue levels of glutathione (GSH/GSSG ratio), lipid peroxides, protein-thiols (P-SH), nitric oxide metabolites, activities of caspase-3 and total superoxide-dismutase (SOD), striatal levels of monoamines and serum levels of hepatic amino-transferases. We observed a decrease of protein-thiols, GSH/GSSG ratio and nitrogen species associated to increased lipid peroxidation in the liver and striatum - but not in the cerebral cortex - of LEC rats, accompanied by dramatic increase in serum amino-transferases and decrease of striatal catecholamines. Conversely, SOD and caspase-3 activity increased consistently only in the cortex of LEC rats. Hence, we assume that enhanced oxidative stress may play a central role in the cell degeneration in WD, at the main sites of copper deposition, with discrete pro-apoptotic conditions developing in distal areas. (c) 2005 Published by Elsevier B.V.

Salvatore MF, Fisher B, Surgener SP, Gerhardt GA, Rouault T. 2005. Neurochemical investigations of dopamine neuronal systems in iron-regulatory protein 2 (IRP-2) knockout mice. Molecular Brain Research 139 (2):341-347.
Abstract: Abnormal iron accumulations are frequently observed in the brains of patients with Parkinson's disease and in normal aging. Iron metabolism is regulated in the CNS by iron regulatory proteins (IRP-1 and IRP-2). Mice engineered to lack IRP-2 develop abnormal motoric behaviors including tremors at rest, abnormal gait, and bradykinesia at middle to late age (18 to 24 months). To further characterize the dopamine (DA) systems of IRP-2 -/- mice, we harvested CNS tissue from age-matched wild type and IRP-2 -/- (16-19 months) and analyzed the protein levels of tyrosine hydroxylase (TH), dopamine transporter (DAT), vesicular monoamine transporter (VMAT2), and DA levels in dorsal striatum, ventral striatum (including the core and shell of nucleus accumbens), and midbrain. We further analyzed the phosphorylation of TH in striatum at serine 40, serine 3 1, and serine 19. In both dorsal and ventral striatum of IRP-2 knockout mice, there was a 20-25% loss of TH protein and accompanied by a similar to 50% increase in serine 40 phosphorylation above wild-type levels. No change in serine 31 phosphorylation was observed. In the ventral striatum, there was also a significant loss (similar to 40%) of DAT and VMAT2. Levels of DA were decreased (similar to 20%) in dorsal striatum, but turnover of DA was also elevated (similar to 30%) in dorsal striatum of IRP-2 -/- mice. We conclude that iron misregulation associated with the loss of IRP-2 protein affects DA regulation in the striatum. However, the modest loss of DA and DA-regulating proteins does not reflect the pathology of PD or

animal models of PD. Instead, these observations support that the IRP-2 -/- genotype may enable neurobiological events associated with aging. (c) 2005 Elsevier B.V. All rights reserved.

Rojas P, Franco-Perez JE, Rojas C, Rojas-Castaneda J, Ebadi M, Fernandez-Valverde F, Serrano-Garcia N. 2005. Reduction of zinc-positive terminal fields in striatum of mouse after 1-methyl-4-phenylpyridinium neurotoxicity. Neurotoxicology 26(6):959-968.
Abstract: Zinc is an essential trace element in the central nervous system and is located in three distinct pools: free zinc, vesicular zinc and protein-bound zinc. Zinc may serve as an endogenous neuromodulator and has been associated with neuropathologies. This study was undertaken to determine whether levels of vesicular zinc in neuronal terminals would decrease in response to the dopaminergic neurotoxin 1-methyl-4-phenylpyridinium (MPP+). Adult male C-57 black mice were injected with MPP+ (0.72 mg/kg) into their right lateral ventricle. All animals were killed at 1, 2, 24 h and 7 days after MPP+ or saline administration. The brains were stained for zinc sulfides and the density of zinc-positive terminal fields was evaluated after MPP+ administration. The relative optical density analysis of zinc-positive terminal fields showed significant decreases in the striatum at 1, 2 and 24 h (24, 18 and 14%, respectively, versus control) and ventricular epithelium (1, 2, 24 h and 7 days). The hippocampus showed increase in the stratum oriens and stratum radiatum at different times. MPP+ administration reduced dopamine levels at 24 h and 7 days (36 and 40%, respectively, versus control) as a result of the neurotoxic action of MPP+. The decrease of zinc-positive neuronal terminal fields in the striatum after MPP+ administration is most likely due to a neuronal release of vesicular zinc in response to its dopaminergic neurotoxicity. (c) 2005 Elsevier Inc. All rights reserved.

Rocchitta G, Migheli R, Mura MP, Grella G, Esposito G, Marchetti B, Miele E, Desole MS, Miele M, Serra PA. 2005. Signaling pathways in the nitric oxide and iron-induced dopamine release in the striatum of freely moving rats: Role of extracellular Ca2+ and L-type Ca2+ channels. Brain Res 1047(1): 18-29.
Abstract: We showed previously that exogenous iron potentiated nitric oxide (NO) donor-induced release of striatal dopamine (DA) in freely moving rats, using microdialysis. In this study, the increase in dialysate DA induced by intrastriatal infusion of the NO-donor 3-morpholinosydnonimine (SIN-1, 1.0 mM for 180 min) was scarcely affected by Ca2+ omission. N-methyl-D-glucamine dithiocarbamate (MGD) is a thiol compound whose NO trapping activity is potentiated by iron(II). Intrastriatal co-infusion of MGD either alone or associated with iron(II), however, potentiated SIN-I-induced increases in dialysate DA. In contrast, co-infusion of the NO trapper 4-(carboxyphenyl)4,4,5,5-tetramethylimidazole- 1-oxyl 3-oxide (carboxy-PTIO) significantly attenuated the increase in dialysate DA induced by SIN-1 (5.0 mM for 180 min). SIN-1 +MGD+iron(II)-induced increases in dialysate DA were inhibited by Ca2+ omission or co-infusion of either deferoxamine or the L-type (Ca-v 1.1 - 1.3) Ca2+ channel inhibitor nifedipine; in contrast, the increase was scarcely affected by co-infusion of the N-type (Ca-v 2.2) Ca2+ channel inhibitor omega-conotoxin GVIA. These results demonstrate that exogenous NO-induced release of striatal DA is independent on extracellular Ca2+; however, in presence of the NO trapper MGD, NO may preferentially react with either endogenous or exogenous iron to form a complex which releases striatal DA with an extracellular Ca2+-dependent and nifedipine-sensitive mechanism. (c) 2005 Elsevier B.V. All rights reserved.

Reaney SH, Smith DR. 2005. Manganese oxidation state mediates toxicity in PC12 cells. Toxicol Appl Pharmacol 205(3):271-281.
Abstract: The role of the manganese (Mn) oxidation state on cellular Mn uptake and toxicity is not well understood. Therefore, undifferentiated PC12 cells were exposed to 0-200 mu M Mn(II)-chloride or Mn(III)-pyrophosphate for 24 h, after which cellular manganese levels were measured along with measures of cell viability, function, and cytotoxicity

(trypan blue exclusion, medium lactate dehydrogenase (LDH), 8-isoprostanes, cellular ATP, dopamine, serotonin, H-ferritin, transferrin receptor (TfR), Mn-superoxide dismutase (MnSOD), and copper-zinc superoxide dismutase (CuZnSOD) protein levels). Exposures to Mn(III) > 10 mu M produced 2- to 5-fold higher cellular manganese levels than equimolar exposures to Mn(II). Cell viability and ATP levels both decreased at the highest Mn(II) and Mn(III) exposures (150-200 mu M), while Mn(III) exposures produced increases in LDH activity at lower exposures (>= 50 mu M) than did Mn(II) (200 mu M only). Mn(II) reduced cellular dopamine levels more than Mn(III), especially at the highest exposures (50% reduced at 200 mu M Mn(II)). In contrast, Mn(III) produced a > 70% reduction in cellular serotonin at all exposures compared to Mn(II). Different cellular responses to Mn(II) exposures compared to Mn(III) were also observed for H-ferritin, TfR, and MnSOD protein levels. Notably, these differential effects of Mn(II) versus Mn(III) exposures on cellular toxicity could not simply be accounted for by the different cellular levels of manganese. These results suggest that the oxidation state of manganese exposures plays an important role in mediating manganese cytotoxicity. (c) 2004 Elsevier Inc. All rights reserved.

Rasia RM, Bertoncini CW, Marsh D, Hoyer W, Cherny D, Zweckstetter M, Griesinger C, Jovin TM, Fernandez CO. 2005. Structural characterization of copper(II) binding to alpha-synuclein: Insights into the bioinorganic chemistry of Parkinson's disease. Proc Natl Acad Sci U S A 102(12): 4294-4299.
Abstract: The aggregation of alpha-synuclein (AS) is characteristic of Parkinson's disease and other neurodegenerative synucleinopathies. We demonstrate here that Cu(II) ions are effective in accelerating AS aggregation at physiologically relevant concentrations without altering the resultant fibrillar structures. By using numerous spectroscopic techniques (absorption, CD, EPR, and NMR), we have located the primary binding for Cu(II) to a specific site in the N terminus, involving His-50 as the anchoring residue and other nitrogen/oxygen donor atoms in a square planar or distorted tetragonal geometry. The carboxylate-rich C terminus, originally thought to drive copper binding, is able to coordinate a second Cu(II) equivalent, albeit with a 300-fold reduced affinity. The NMR analysis of AS-Cu(II) complexes reveals the existence of conformational restrictions in the native state of the protein. The metallobiology of Cu(II) in Parkinson's disease is discussed by a comparative analysis with other Cu(II)-binding proteins involved in neurodegenerative disorders.

Rainero I, Rivoiro C, Rubino E, Milli V, Valfre W, De Martino P, Lo Giudice R, Angilella G, Savi L, Gallone S, Pinessi L. 2005. Prevalence of HFE (hemochromatosis) gene mutations in patients with cluster headache. Headache 45(9):1219-1223.
Abstract: Objective.-To evaluate whether polymorphisms of the HFE gene would modify the occurrence and the clinical features of cluster headache (CH). Background.-Recent studies suggested that iron metabolism may be involved in the pathophysiology of primary headaches. The HFE gene encodes for a protein that modulates iron absorption. Mutations in this gene are responsible for toxic iron overload in several body organs. Methods.-Genomic DNA was extracted from 109 CH patients and 211 age and sex-matched healthy controls and genotyped for the C282Y and H63D mutations of the HFE gene. Allele and genotype frequencies of the HFE gene were compared between cases and controls. The clinical characteristics of the disease were compared according to the different HFE gene genotypes. Results.-No C282Y mutation was found in both cases and controls. The prevalence of the H63D mutation was nearly identical in cases and controls. The four patients carrying the HFE D63D genotype showed a significantly ($P < .001$) later age at onset of the disease in comparison with both H63H and H63D patients. The remaining clinical characteristics of the disease did not significantly differ in the presence or absence of the H63D mutation. Conclusion.-Our data do not support the hypothesis that genetic variations within the HFE gene are associated with CH. However, the HFE gene may influence the disease phenotype and may

be regarded as a disease modifier gene.

Raicevic N, Mladenovic A, Perovic M, Harhaji L, Miljkovic D, Trajkovic V. 2005. Iron protects astrocytes from 6-hydroxydopamine toxicity. Neuropharmacology 48(5):720-731.
Abstract: The role of iron in 6-hydroxydopamine (6-OHDA) toxicity towards astrocytes was investigated in vitro using rat primary astrocytes, rat astrocytoma cell line C6, and human astrocytoma cell line U251. The assessment of mitochondrial respiration or lactate dehydrogenase release has shown a dose-dependent decrease in the viability of astrocytes treated with 6-OHDA, which coincided with DNA fragmentation and the changes in cellular morphology. This was a consequence of the oxidative stress mediated by 6-OHDA autoxidation products hydrogen peroxide, superoxide anion, and hydroxyl radical. Both FeSO4 and FeCl3 markedly alleviated detrimental effects of 6-OHDA treatment, while MgSO4 was without effect. The protective action of iron was neutralized by a membrane-permeable iron chelator o-phenanthroline, which also augmented astrocyte killing in the absence of exogenous iron. The mechanisms responsible for iron-mediated protection of astrocytes did not involve interference with either 6-OHDA autoxidation, hydrogen peroxide toxicity, or 6-OHDA-induced activation of extracellular signal-regulated kinase. Finally, the addition of iron potentiated and its chelation blocked 6-OHDA toxicity towards neuronal PC12 cells, suggesting the opposite roles for this transition metal in regulating the survival of astrocytes and dopaminergic neurons. (c) 2005 Elsevier Ltd. All rights, reserved.

Racette BA, Antenor JA, Mcgee-Minnich L, Moerlein SM, Videen TO, Kotagal V, Perlmutter JS. 2005. [F-18]FDOPA PET and clinical features in parkinsonism due to manganism. Mov Disord 20(4):492-496.
Abstract: Manganese exposure reportedly causes a clinically and pathophysiologically distinct syndrome from idiopathic Parkinson's disease (PD). We describe the clinical features and results of positron emission tomography with 6-[F-18]fluorodopa ([F-18]FDOPA PET) of a patient with parkinsonism occurring in the setting of elevated blood manganese. The patient developed parkinsonism associated with elevated serum manganese from hepatic dysfunction. [F-18]FDOPA PET demonstrated relatively symmetric and severely reduced [F-18]FDOPA levels in the posterior putamen compared to controls. The globus pallidum interna had increased signal on T1-weighted magnetic resonance imaging (MRI) images. We conclude that elevated manganese exposure may be associated with reduced striatal [F-18]FDOPA uptake, and MRI may reveal selective abnormality within the internal segment of the pallidum. This case suggests that the clinical and pathophysiological features of manganese-associated parkinsonism may overlap with that of PD.

Quintana M, Klouda AD, Ochsenkuhn-Petropoulou M, Michalke B. 2005. Size characterization of manganese species from liver extracts using size exclusion chromatography inductively coupled plasma mass spectrometry. Anal Chim Acta 554(1-2):130-135.
Abstract: Increased Mn levels are known to damage the central nervous system, resulting in motoric abnormalities and psychic disorder and finally resulting even in symptoms similar to Parkinson's disease. Monomethyl-Mn-pentadienyl-tricarbonyl is used as anti-knock agent and consequently Mn compounds are exhausted into air from automobiles. With additional inhalative Mn exposure, finally an Mn overflow of the liver is known, resulting in increased Mn transport to other organs, predominantly to the brain. Specific Mn-species then seem to be generated in liver, however, their speciation is still not investigated. This paper focuses on experiments to get more information on Mn species with respect to a size characterization of the Mn species in liver. Liver extracts were analyzed using a mass calibrated size exclusion chromatography (SEC) column being coupled to inductively coupled plasma mass spectrometry (ICP-MS) detection. As an important prerequisite, the stability of Mn species in the liver extracts during storage was investigated as well. It turned out that short term storage of the extract (under Ar atmosphere) at 4 degrees C

seems to be best suited. Storage for several days even at -20 degrees C demonstrated already considerable changes in species pattern. Further investigations focused on improvements in detection during hyphenation using dynamic reaction cell (DRC) technology for Mn detection. The signal to noise (S/N) ratio was increased up to a factor of 15 when using DRC technology compared to conventional ICP-MS without DRC. The analysis of liver extracts with a mass calibrated SEC column coupled to inductively coupled plasma-dynamic reaction cell-mass spectrometry (ICP-DRC-MS) showed Mn associated to ca. 36% to a peak covering a mass range 100-260 kDa, approximately 9% was found in a peak having the mass range 37-77 kDa, 46% in a peak having the mass range 13-36 kDa and ca. 7% in the low molecular mass (LMM) range. (c) 2005 Elsevier B.V. All rights reserved.

Qian YC, Zheng Y, Ramos KS, Tiffany-Castiglioni E. 2005. The involvement of copper transporter in lead-induced oxidative stress in astroglia. Neurochem Res 30(4):429-438.
Abstract: Lead (Pb), depositing primarily in astroglia in the brain, is a well-known neurotoxicant and a risk factor for neurologic disorders. Pb has been reported to induce oxidative stress by probably the disturbance of copper (Cu) homeostasis in astroglia. Thus, we hypothesized that Pb-induced oxidative stress is initiated by interfering with Cu transporter in astroglia. In this study, we observed Pb-induced oxidative stress as indicated by reactive oxygen species (ROS) augmentation and GRP78 and GRP94 protein induction, and it was parallel to Cu accumulation intracellularly by Pb. To further address Cu transporter as a potential Pb target, a heavy metal-binding (HMB) domain of Cu-transporting ATPase (Atp7a) was overexpressed and purified. Evidence showed that one molecule of HMB chelated 11 Pb ions or seven Cu ions and that Pb competed with Cu for binding to HMB. These findings suggest that Pb-induced oxidative stress results from the impairment of Cu metabolism by Pb targeting of Atp7a.

Qian YC, Zheng Y, Abraham L, Ramos KS, Tiffany-Castiglioni E. 2005. Differential profiles of copper-induced ROS generation in human neuroblastoma and astrocytorna cells. Molecular Brain Research 134(2):323-332.
Abstract: To determine neuronal and glial responses to copper (Cu) elevation in the CNS, human neuroblastoma and astrocytoma cells were used to compare their responses to Cu in terms of reactive oxygen species (ROS) generation and expression of enzymes responsible for anti-oxidation. Astrocytoma cells, not neuroblastoma cells, were responsive to Cu and Cu elevation was associated with ROS generation. Intracellular Cu levels as determined by inductively coupled plasma-mass spectrometry (ICP-MS), and expression levels of copper-transporting AtPase (ATP7A) and human copper transporter 1 (hCtr1) as detected by quantitative reverse transcription-polymerase chain reaction (RT-PCR), were comparable in both cell lines. Differences in Cu-induced ROS between two cell lines paralleled superoxide dismutase (SOD)-catalase expression as detected by Western blot analysis. Copper, zinc-SOD (Cu,Zn-SOD) and catalase protein levels were upregulated by Cu in neuroblastoma cells while Cu,Zn-SOD was down-regulated by Cu and catalase level wag not changed in astrocytoma cells. Manganese-SOD (Mn-SOD) was not responsive to Cu in either cell line. Furthermore, 78-kDa glucose-regulated protein aggregation and upregulation were observed in Cu-treated astrocytoma cells, but not neuroblastoma cells. These data suggest that neurons use the SOD-catalase system to scavenge Cu-induced ROS while glia rely on the endoplasmic reticulum stress response to compensate for the reduction of ROS scavenging capacity. (c) 2004 Elsevier B.V. All rights reserved.

Pountney DL, Voelcker NH, Gai WP. 2005. Annular alpha-synuclein oligomers are potentially toxic agents in alpha-synucleinopathy. Hypothesis. Neurotoxicity Research 7(1-2):59-67.
Abstract: Recently, we demonstrated that soluble 30-50 nm-sized annular alpha-synuclein oligomers are released by mild detergent treatment from

glial cytoplasmic inclusions (GCIs) purified from multiple system atrophy brain tissue (Pountney et al, J. Neurochem. 90:502, 2004). Dynamic antibody recognition imaging using a specific anti-alpha-synuclein antibody confirmed that the annular structures were positive for alpha-synuclein. This showed that pathological alpha-synucleinopathy aggregates can be a source of annular alpha-synuclein species. In contrast to pathological alpha-synuclein, recombinant a-synuclein yielded only spherical oligomers after detergent treatment, indicating a greater propensity of the pathological protein to form stable annular oligomers. In vitro, we found that Cal(2+) binding to monomeric a-synuclein, specifically amongst a range of different metal ions, induced the rapid formation of annular oligomers (Lowe et al., Protein Sci., 13:3245, 2004). Hence, alpha-synuclein speciation may also be influenced by the intracytoplasmic Cal(2 +) concentration. We also showed that annular alpha-synuclein oligomers can nucleate filament formation. We hypothesize that soluble alpha-synuclein annular oligomers may be cytotoxic species, either by interacting with cell membranes or components of the ubiquitin proteasome system. The equilibrium between alpha-synuclein species may be influenced by intracellular Cal(2+) status, interaction with lipid vesicles or other factors.

Platonova NA, Barabanova SV, Povalikhin RG, Tsymbalenko NV, Danilovskii MA, Voronina OV, Dorokhova II, Puchkova LV. 2005. In vivo expression of copper-transporting proteins in rat brain regions. Biology Bulletin 32(2): 108-120.
Abstract: Expression of two copper-transporting P1-type ATPases (ATP7A and ATP7B), the CTR1 protein, a high-affinity copper transporter, and ceruloplasmin (Cp), a copper-containing ferroxidase was studied. The level of mRNA of these proteins was determined by RT-PCR analysis, the distribution of polypeptides encoded by these genes was determined by immunoblotting, and the type of cells expressing these genes was identified immunohistochemically. It was found that the major product of Cp gene in the brain is the cell membrane-bound Cp. Secretory Cp, whose molecule contains the greatest number of weakly associated copper atoms, is synthesized in the choroid plexus. CTR1 rnRNA is evenly distributed in the brain; however, its content is twice higher in the vascular plexus. The Atp7a gene is active in all brain regions, whereas the Atp7b gene is active only in the hypothalamus. The membrane-bound Cp is expressed in glial cells of all types and in ependyma cells. ATP7B and ATP7A are expressed predominantly in ependymyocytes and neurons, respectively. The organization of copper transport in mammalian brain is discussed.

Piruat JI, Lopez-Barneo J. 2005. Oxygen tension regulates mitochondrial DNA-encoded complex I gene expression. J Biol Chem 280(52):42676-42684.
Abstract: Oxygen is a major regulator of nuclear gene expression. However, although mitochondria consume almost all of the O-2 available to the cells, little is known about how O-2 tension influences the expression of the mitochondrial genome. We show in O-2-sensitive excitable rat PC12 cells that, among the mtDNA- encoded genes, hypoxia produced a specific down-regulation of the transcripts encoding mitochondrial complex I NADH dehydrogenase (ND) subunits, particularly ND4 and ND5 mRNAs and a stable mRNA precursor containing the ND5 and cytochrome b genes. This unprecedented effect of hypoxia was fast (developed in < 30 min) and fairly reversible and occurred at moderate levels of hypoxia (O-2 tensions in the range of 20 - 70 mm Hg). Hypoxic down-regulation of the mitochondrial complex I genes was paralleled by the reduction of complex I activity and was retarded by iron chelation, suggesting that an iron-dependent post- transcriptional mechanism could regulate mitochondrial mRNA stability. It is known that cell respiration is under tight control by the amount of proteins in mitochondrial complexes of the electron transport chain. Therefore, regulation of the expression of the mitochondrial (mtDNA)-encoded complex I subunits could be part of an adaptive mechanism to adjust respiration rate to the availability of O-2 and to induce fast adaptive changes in hypoxic cells.

Petersen RB, Siedlak SL, Lee HG, Kim YS, Nunomura A, Tagliavini F, Ghetti B,

Cras P, Moreira PI, Castellani RJ, Guentchev M, Budka H, Ironside JW, Gambetti P, Smith MA, Perry G. 2005. Redox metals and oxidative abnormalities in human prion diseases. Acta Neuropathol (Berl) 110(3): 232-238.
Abstract: Prion diseases are characterized by the accumulation of diffuse and aggregated plaques of protease-resistant prion protein (PrP) in the brains of affected individuals and animals. Whereas prion diseases in animals appear to be almost exclusively transmitted by infection, human prion diseases most often occur sporadically and, to a lesser extent, by inheritance or infection. In the sporadic cases (sporadic Creutzfeld-Jakob disease, sCJD), PrP-containing plaques are infrequent, whereas in transmitted (variant CJD) and inherited (Gerstmann-Straussler-Scheinker Syndrome) cases, plaques are a usual feature. In the current study, representative cases from each of the classes of human prion disease were analyzed for the presence of markers of oxidative damage that have been found in other neurodegenerative diseases. Interestingly, we found that the pattern of deposition of PrP, amyloid-beta, and redox active metals was distinct for the various prion diseases. Whereas 8-hydroxyguanosine has been shown to be increased in sCJD, and inducible NOS is increased in scrapie-infected mice, well-studied markers of oxidative damage that accumulate in the lesions of other neurodegenerative diseases (such as Alzheimer's disease, progressive supranuclear palsy, and Parkinson's disease), such as heme oxygenase-1 and lipid peroxidation, were not found around PrP deposits or in vulnerable neurons. These findings suggest an important distinction in prion-related oxidative stress, indicating that different neurodegenerative pathways are involved in different prion diseases.

Perez-Martinez L, Jaworski DM. 2005. Tissue inhibitor of metalloproteinase-2 promotes neuronal differentiation by acting as an anti-mitogenic signal. J Neurosci 25(20):4917-4929.
Abstract: Although traditionally recognized for maintaining extracellular matrix integrity during morphogenesis, the function of matrix metalloproteinases (MMPs) and their inhibitors, the tissue inhibitors of metalloproteinases (TIMPs), in the mature nervous system is essentially unknown. Here, we report that TIMP-2 induces pheochromocytoma PC12 cell-cycle arrest via regulation of cell-cycle regulatory proteins, resulting in differentiation and neurite outgrowth. TIMP-2 decreases cyclins B and D expression and increases p21(Cip) expression. Furthermore, TIMP-2 promotes cell differentiation via activation of the cAMP/Rap1/ERK (extracellular signal-regulated kinase) pathway. Expression of dominant-negative Rap1 blocks TIMP-2-mediated neurite outgrowth. Both the cell-cycle arrest and neurite outgrowth induced by TIMP-2 was independent of MMP inhibitory activity. Consistent with the PC12 cell data, primary cultures of TIMP-2 knockout cerebral cortical neurons exhibit significantly reduced neurite length, which is rescued by TIMP-2. These in vitro results were corroborated in vivo. TIMP-2 deletion causes a delay in neuronal differentiation, as demonstrated by the persistence of nestin-positive progenitors in the neocortical ventricular zone. The interaction of TIMP-2 with alpha 3 beta 1 integrin in the cerebral cortex suggests that TIMP-2 promotes neuronal differentiation and maintains mitotic quiescence in an MMP-independent manner through integrin activation. The identification of molecules responsible for neuronal quiescence has significant implications for the ability of the adult brain to generate new neurons in response to injury and neurological disorders, such as Alzheimer's and Parkinson's diseases.

Park J, Yoo CI, Sim CS, Kim HK, Kim JW, Jeon BS, Kim KR, Bang OY, Lee WY, Yi Y, Jung KY, Chung SE, Kim Y. 2005. Occupations and Parkinson's disease: A multi-center case-control study in South Korea. Neurotoxicology 26(1): 99-105.
Abstract: Objective: We performed a hospital based case-control study in South Korea (1) to clarify the role of occupational exposure, and especially manganese (Mn) exposure in the etiology of Parkinson's disease (PD) and (2) to discover the association between any occupations and PD. Methods:

We selected two groups, PD patient group (NI) and controls (N-2). Three hundred sixty-seven consecutive outpatients with PD (177 men, 190 women) and 309 controls were interviewed about life style, past history, family history, education level, and occupational history etc. We employed a range of industrial categories as defined by section (the most broad category) and division (sub-category) of the Korea Standard Industry Code (KSIC) Manual. Along with KSIC, we also used the Korea Standard Classification of Occupations (KSCO) as proxies of occupational exposure. The odds ratios (ORs) and 95% confidence intervals (CA), adjusted for age, sex, smoking status, and education level are presented. Results: As regarding the exposure to hazardous materials, especially Mn, more subjects in the control group than the PD patient group 'have worked in the occupations with potential exposure to Mn ($P < 0.001$). Ever having worked in 'agriculture, hunting, and forestry' section of industry was positively associated with PD (OR 1.88), and 'agriculture production crops (OR 1.96)'division of industry was positively associated with PD. On the other hand, ever having worked in the 'manufacturing (OR 0.56)', 'transportation (OR 0.28)' section of industry, and 'transporting (OR 0.20)' division of industry were negatively associated with PD. 'Drivers (OR 0.13)'division of occupation also was negatively associated with PD. Conclusions: To our knowledge, this is the first case-control studies to find an inverse relationship between 'transporting' or 'technicians like machinery engineers' as his/her longest job and PD risk. Because of this unexpected finding, our work should be replicated in various populations. (C) 2004 Elsevier Inc. All rights reserved.

Paris I, Martinez-Alvarado P, Perez-Pastene C, Vieira MNN, Olea-Azar C, Raisman-Vozari R, Cardenas S, Graumann R, Caviedes P, Segura-Aguilar J. 2005. Monoamine transporter inhibitors and norepinephrine reduce dopamine-dependent iron toxicity in cells derived from the substantia nigra. J Neurochem 92(5):1021-1032.
Abstract: The role of dopamine in iron uptake into catecholaminergic neurons, and dopamine oxidation to aminochrome and its one-electron reduction in iron-mediated neurotoxicity, was studied in RCSN-3 cells, which express both tyrosine hydroxylase and monoamine transporters. The mean +/- SD uptake of 100 muM (FeCl3)-Fe-59 in RCSN-3 cells was 25 +/- 4 pmol per min per mg, which increased to 28 +/- 8 pmol per min per mg when complexed with dopamine (Fe(III)-dopamine). This uptake was inhibited by 2 muM nomifensine (43% $p < 0.05$), 100 muM imipramine (62% $p < 0.01$), 30 muM reboxetine (71% $p < 0.01$) and 2 mM dopamine (84% $p < 0.01$). The uptake of Fe-59-dopamine complex was Na+, Cl- and temperature dependent. No toxic effects in RCSN-3 cells were observed when the cells were incubated with 100 muM FeCl3 alone or complexed with dopamine. However, 100 muM Fe(III)-dopamine in the presence of 100 muM dicoumarol, an inhibitor of DT-diaphorase, induced toxicity (44% cell death; $p < 0.001$), which was inhibited by 2 muM nomifensine, 30 muM reboxetine and 2 mM norepinephrine. The neuroprotective action of norepinephrine can be explained by (1) its ability to form complexes with Fe3+, (2) the uptake of Fe-norepinephrine complex via the norepinephrine transporter and (3) lack of toxicity of the Fe-norepinephrine complex even when DT-diaphorase is inhibited. These results support the proposed neuroprotective role of DT-diaphorase and norepinephrine.

Paris I, Martinez-Alvarado P, Cardenas S, Perez-Pastene C, Graumann R, Fuentes P, Olea-Azar C, Caviedes P, Segura-Aguilar J. 2005. Dopamine-dependent iron toxicity in cells derived from rat hypothalamus. Chem Res Toxicol 18 (3):415-419.
Abstract: We report a new and specific mechanism for iron-mediated neurotoxicity using RCHT cells, which were derived from rat hypothalamus. RCHT cells exhibit immunofluorescent-positive markers for dopamine beta-hydroxylase and the norepinephrine transporter, NET. In the present study, we observed that iron-induced neurotoxicity in RCHT cells was dependent on (i) formation of an Fe-dopamine complex (100 mu M FeCl3:100 mu M dopamine); (ii) specific uptake of the Fe-dopamine complex into RCHT cells via NET (79 +/- 2 pmol Fe-59/mg/min; $P < 0.05$),

since the uptake of the Fe-59-dopamine complex by the cells was inhibited by 30 mu M reboxetine, a specific NET inhibitor (78% inhibition, P < 0.001); and (iii) intracellular oxidation of dopamine present in the Fe-dopamine complex to aminochrome; (iv) inhibition of DT-diaphorase, since incubation of RCHT cells with 100 mu M Fe-dopamine complex in the presence of 100 mu M dicoumarol, an inhibitor of DT-diaphorase, induced significant cell death (51 +/- 5%; P < 0.061). However, this cell death was reduced by 75% when the cells were incubated in the presence of 30 mu M reboxetine (P < 0.01). No significant cell death was observed when the cells were incubated with 100 mu M dopamine, 100 mu M Fe-Dopamine complex, 100 mu M dicoumarol, or 100 mu M FeCl3 (8.3 +/- 2, 9 +/- 4, 8.5 +/- 3, or 9.7 +/- 2% of control, respectively). ESR studies using the spin trapping agent DMPO showed no formation of hydroxyl radicals when the cells were incubated with 100 mu M FeCl3 alone. However, using the same ESR technique, the formation of hydroxyl radicals and a carbon-centered radical was detected when the cells were incubated with 100 mu M Fe-dopamine complex in the presence of 100 mu M dicoumarol. These studies suggest that iron can induce cell toxicity by a mechanism that requires the formation and NET-mediated uptake of an Fe-dopamine complex, ultimately resulting in the intracellular formation of reactive species.

Pappert EJ. 2005. Toxin-induced movement disorders. Neurol Clin 23(2):429-+.
Abstract: Most instances of confirmed toxin-induced movement disorders show lesions on CT and MRI scans of cortical or subcortical structures. A common underlying element in these toxin-induced syndromes is the development of lesions primarily in the pallidum and striatum. Because many toxins result in lesions affecting these structures, a selective vulnerability to hypoxic or metabolic insults has long been postulated. The susceptibility of these structures may related to a number of factors including the pattern of oxidative metabolism, heavy metal concentration, vascular perfusion, and neuronal innervation. In addition to causing disability, certain neurotoxins have led to a better understanding of human disease through the development of research models.

Pande MBS, Nagabhushan P, Hegde ML, Rao TSS, Rao KSJ. 2005. An algorithmic approach to understand trace elemental homeostasis in serum samples of Parkinson disease. Comput Biol Med 35(6):475-493.
Abstract: A classical problem in neurological disorders is to understand the progression of disorder and define the trace elements (metals) which play a role in deviating a sample from normal to an abnormal state, which implies the need to create a reference knowledge base (KB) employing the control samples drawn from normal/healthy set in the context of the said neurological disorder, and in sequel to analytically understand the deviations in the cases of disorders/abnormalities/unhealthy samples. Hence building up a computational model involves mining the healthy control samples to create a suitable reference KB and designing an algorithm for estimating the deviation in case of unhealthy samples. This leads to realizing an algorithmic cognition-recognition model, where the cognition stage establishes a reference model of a normal/healthy class and the recognition stage involves discriminating whether a given test sample belongs to a normal class or not. Further if the sample belongs to a specified reference base (normal) then the requirement is to understand how strong the affiliation is, and if otherwise (abnormal) how far away the sample is from the said reference base. In this paper, an exploratory data analysis based model is proposed to carry out such estimation analysis by designing distribution and parametric models for the reference base. Further, the knowledge of the reference base in case of the distribution model is expressed in terms of zones with each zone carrying a weightage factor. Different distance measures are utilized for the subsequent affiliation analysis (City block with distribution model and Doyle's with Parametric model). Results of an experimental study based on the database of trace elemental analysis in human serum samples from control and Parkinson's neurological disorder are presented to corroborate the performance of the computational algorithm. (C) 2004 Published by

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Ozekmekci S, Ertan S, Kiziltan G, Apaydin H, Djaksybaeva D, Sayilir I. 2005. Progressive parkinsonism in a welder due to manganese intoxication. Mov Disord 20:S106.

Oyanagi K. 2005. The nature of the parkinsonism-dementia complex and amyotrophic lateral sclerosis of Guam and magnesium deficiency. Parkinsonism & Related Disorders 11:S17-S23.
Abstract: The parkinsonism-dementia complex (PDC) and amyotrophic lateral sclerosis (ALS) were the fatal neurological diseases, showing very high incidence during 1950-1970 and dramatic decrease after 1970 on Guam. Through the research, the present author insisted that: (1) NFTs in Guam ALS patients are merely a background feature widely dispersed in the population, (2) Guam ALS and PDC are basically different diseases, and (3) Guam ALS occurs initially as classic ALS. As pathogeneses of the diseases, intake of low calcium (Ca) and magnesium (Mg) and high aluminum water and of some plant excitatory neurotoxin has been speculated. To elucidate the pathogenesis. the author per-formed an experiment exposing rats to low Ca and/or Mg intake for two generations, so as to follow the actual way of human living on the island, since several generations live continuously in the same environment. The study indicates that continuous low Mg intake for two generations induces exclusive loss of dopaminergic neurons in in rats. and may support the Mg hypothesis in the pathogenesis of PDC of Guam. (C) 2005 Elsevier Ltd. All rights reserved.

Ohtake T, Negishi K, Okamoto K, Oka M, Maesato K, Moriya H, Kobayashi S. 2005. Manganese-induced parkinsonism in a patient undergoing maintenance hemodialysis. Am J Kidney Dis 46(4):749-753.
Abstract: We report a rare case of manganese (Mn)-induced parkinsonism in a patient on maintenance hemodialysis therapy who complained of gait disturbance and dysarthria. His symptoms and abnormal magnetic resonance imaging (MRI) findings of the brain were thought to be caused, at least in part, by long-term ingestion of a health supplement (Chlorella extract) that contained 1.7 mg of Mn in the usual daily dose. Elevated serum and cerebrospinal fluid Mn levels were detected, and brain MRI showed areas of abnormal intensity in the bilateral basal ganglia (low intensity on T1-weighted images and high intensity on T2-weighted images). Edetic acid infusion therapy dramatically improved the MRI abnormalities, after which his symptoms gradually improved 4 months later.

Novakovic KE, Villemagne VL, Rowe CC, Masters CL. 2005. Rare genetically defined causes of dementia. Int Psychogeriatr 17:S149-S194.
Abstract: Several genetic disorders, though rare, are associated or present with dementia. Developments in the field of genetics are contributing to clarify and expand our knowledge of the complex physiopathological mechanisms leading to neurodegeneration and cognitive decline. Disorders associated with misfolded and aggregated proteins and lipid, metal or energy metabolism are examples of the multifarious disease processes converging in the clinical features of dementia, either as its predominant feature, as in cases of Alzheimer's disease (AD) or frontotemporal dementia (FTD), or as part of a cohort of accompanying or late-developing symptoms, as in Parkinson's disease (PD) or amyotrophic lateral sclerosis with dementia (ALS-D). Awareness of these disorders, allied with recent advances in genetic, biochemical and neuroimaging techniques, may lead to early diagnosis, successful treatment and better prognosis.

Niu PY, Niu Q, Zhang QL, Wang LP, He SC, Wu TC, Conti P, Di Gioacchino M, Boscolo P. 2005. Aluminum impairs rat neural cell mitochondria in vitro. International Journal of Immunopathology and Pharmacology 18(4): 683-689.
Abstract: Exposure to aluminum has been reported to lead to neurotoxicity. Mitochondria are important organelles involved in maintaining cell function. This study investigates the effect of aluminum on

mitochondria in rat neural cells. The ultrastructure of mitochondria was observed, and the cell death rate (CDR), reactive oxygen species (ROS), mitochondrial membrane potential (MMP) and 3-[4,5demethyl-2-thiazalyl]-2,-5diphenyl-2H-tetrazolium bromide (MTT) were measured to investigate the effect of aluminum on the mitochondrial structure and its function in neural cells. Results observed from the mitochondrial ultrastructure show that aluminum may impair the mitochondrial membrane and cristae. Increased CDR, enhanced ROS, decreased MMP, and decreased enzyme activity in mitochondria were observed in the AI-exposed neurons (100 - 500 mu M). The present study demonstrates that alteration in the mitochondrial structure and function plays an important role in neurotoxic mechanisms induced by aluminum.

Nicolaus BJR. 2005. A critical review of the function of neuromelanin and an attempt to provide a unified theory. Med Hypotheses 65 (4):791-796. Abstract: This paper provides a critical review of the numerous and various biological functions so far attributed to neuromelanin and an attempt to provide a unified theory based on the peculiar physical and chemical properties of the black particle (the neuromelanin cage). It is stressed that neuromelanin is not homogeneous, as is commonly accepted, but is made up of different substrate specific black pigments formed by the oxidation of o.diphenots or other oxygenated precursors (substantia nigra melanin, locus coeruleus melanin, retinal pigmented epithelium or ocular melanin, inner-ear melanin, and so on). Ocular melanin is believed to protect the eye by trapping metals and free radicals. The paper shows that this unconfirmed mechanism is a rather fortuitous irreversible molecular accident, which at times may prove itself deleterious. Albinism often leads to deafness in animals, indicating a genetic correlation. These two conditions appear to be correlated at a molecular level to eye/ear pigmentation and suggest verifying this hypothesis in normal and albino human individuals. Skin and ocular melanin are chemically different. However, they are both involved in light absorption/dissipation. The black particle structure (melanin cage) is believed to be fundamental to this process because there is a common bioelectric mechanism. The latter is worth of further investigation. It is also proposed checking how ocular melanin dissipates the excessive absorbed light (as heat or as current?). It has been claimed that inner-ear melanin mutes acoustic waves. This paper suggests investigating the underlying mechanism and also studying whether this pigment is bioelectrically involved in audiology. According to numerous authors, substantia nigra melanin is only biological garbage. This view is rejected, and it is stressed that intracellular melanogenesis is a fundamental and genetically controlled physiological process. It has been repeatedly claimed that the binding of iron, heavy metals, free radicals and harmful chemicals by substantia nigra melanin is fundamental to body detoxification/protection. Presumably, such irreversible and generic binding mechanisms have no physiological foundation; it is suggested the alternative that, substantia nigra melanin acts as semiconductor, transmitting and modulating nervous impulses, in a reversible way. In fact, substantia nigra melanin is absent or significantly scarce in two conditions of life in which the coordination of movement is either inefficient (newborn babies) or strongly compromised (Parkinson). To check this assumption, further investigation of nucleus caudatus, putomen, globus pallidus, substantia nigra pars compacta and reticulata, nucleus hypothalamicus is recommended. (c) 2005 Elsevier Ltd. All rights reserved.

Nguyen T, Hamby A, Massa SM. 2005. Clioquinol down-regulates mutant huntingtin expression in vitro and mitigates pathology in a Huntington's disease mouse model. Proc Natl Acad Sci U S A 102(33):11840-11845. Abstract: In investigating the role of metal ions in the pathogenesis of Huntington's disease, we examined the effects of clioquinol, a metal-binding compound currently in clinical trials for Alzheimer's disease treatment, on mutant huntingtin-expressing cells. We found that PC12 cells expressing polyglutarnine-expanded huntingtin exon 1 accumulated less mutant protein and showed decreased cell death when treated with clioquinol. This effect was polyglutamine-length-specific and did not alter

mRNA levels or protein degradation rates. Clioquinol treatment of transgenic Huntington's mice (R6/2) improved behavioral and pathologic phenotypes, including decreased huntingtin aggregate accumulation, decreased striatal atrophy, improved rotarod performance, reduction of weight loss, normalization of blood glucose and insulin levels, and extension of lifespan. Our results suggest that clioquinol is a candidate therapy for Huntington's disease and other polyglutamine-expansion diseases.

Munch C, Rosenbohm A, Sperfeld AD, Uttner I, Reske S, Krause BJ, Sedlmeier R, Meyer T, Hanemann CO, Stumm G, Ludolph AC. 2005. Heterozygous R1101K mutation of the DCTN1 gene in a family with ALS and FTD. Ann Neurol 58(5):777-780.
Abstract: A heterozygous R1101K mutation of the p150 subunit of dynactin (DCTN1) is reported in a family with amyotrophic lateral sclerosis (ALS) and co-occurrence of frontotemporal dementia (FTD). Two members of our kindred were affected with motor neuron disease and two with dementia in an autosomal dominant pattern of inheritance. We excluded the involvement of the AILS and FTD-linked genes for copper/zinc superoxide dismutase (SOD1) and tau. The R1101K sequence alteration of the DCTN1 gene may predispose subjects to ALS and FTD.

Moreira PI, Siedlak SL, Aliev G, Zhu X, Cash AD, Smith MA, Perry G. 2005. Oxidative stress mechanisms and potential therapeutics in Alzheimer disease. J Neural Transm 112(7):921-932.
Abstract: Oxidative damage of biological macromolecules is a hallmark of most neurodegenerative disorders such as Alzheimer, Parkinson and diffuse Lewy body diseases. Another important phenomenon involved in these disorders is the alteration of iron and copper homeostasis. Data from the literature support the involvement of metal homeostasis in mitochondrial dysfunction, protein alterations and nucleic acid damage which are relevant in brain function and consequently, in the development of neurodegenerative disorders. Although alterations in transition metal homeostasis, redox activity, and localization are well documented, it must be determined how alterations of specific copper- and iron-containing metalloenzymes are also involved in Alzheimer disease. The clarification of these phenomena can open a new window for understanding the mechanisms underlying neurodegeneration and, consequently, for the development of new therapeutic strategies such as gene therapy and new pharmaceutical formulations with antioxidant and chelating properties.

Morawski M, Meinecke C, Reinert T, Dorffel AC, Riederer P, Arendt T, Butz T. 2005. Determination of trace elements in the human substantia nigra. Nuclear Instruments & Methods in Physics Research Section B-Beam Interactions With Materials and Atoms 231(Sp. Iss. Si):224-228.
Abstract: "The gain in brain is mainly in the stain" was long time a key sentence for research in neurodegenerative disease. However, for a quantification of the element concentrations (especially iron) in brain tissue, standard staining methods are insufficient. Advanced physical methods allow a quantitative elemental analysis of brain tissue. The sophisticated ion beam analysis provides a quantitative determination of elemental concentrations with a subcellular spatial resolution using a scanning proton beam focussed down to below 1 mu m that induces characteristic X-rays in the specimen (PIXE - particle induced X-ray emission).

Mocchegiani E, Bertoni-Freddari C, Marcellini F, Malavolta M. 2005. Brain, aging and neurodegeneration: Role of zinc ion availability. Prog Neurobiol 75(6): 367-390.
Abstract: Actual fields of research in neurobiology are not only aimed at understanding the different aspects of brain aging but also at developing strategies useful to preserve brain compensatory capacity and to prevent the onset of neurodegenerative diseases. Consistent with this trend much attention has been addressed to zinc metabolism. In fact, zinc acts as a neuromodulator at excitatory synapses and has a considerable role in the

stress response and in the functionality of zinc-dependent enzymes contributing to maintaining brain compensatory capacity. In particular, the mechanisms that modulate the free zinc pool are pivotal for safeguarding brain health and performance. Alterations in zinc homeostasis have been reported in Parkinson's and Alzheimer's disease as well as in transient forebrain ischemia, seizures and traumatic brain injury, but little is known regarding aged brain. There is much evidence that that age-related changes, frequently associated to a decline in brain functions and impaired cognitive performances, could be related to dysfunctions affecting the intracellular zinc ion availability. A general agreement emerges from studies of humans' and rodents' old brains about an increased expression of metallothionein (NIT) isoforms I and II, but dyshomogenous results are reported for MT-III, and it is still uncertain whether these proteins maintain in aging the protective role, as it occurs in adult/young age. At the same time, there is considerable evidence that amyloid-P deposition in Alzheimer's disease is induced by zinc, but the pathological significance and the causes of this phenomenon are still an open question. The scientific debate on the role of zinc and of some zinc-binding proteins in aging and neurodegenerative disorders, as well as on the beneficial effect of zinc supplementation in aged brain and neurodegeneration, is extensively discussed in this review. (c) 2005 Elsevier Ltd. All rights reserved.

Mikhaylova A, Davidson M, Toastmann H, Channell JET, Guyodo Y, Batich C, Dobson J. 2005. Detection, identification and mapping of iron anomalies in brain tissue using X-ray absorption spectroscopy. Journal of the Royal Society Interface 2(2):33-37.
Abstract: This work describes a novel method for the detection, identification and mapping of anomalous iron compounds in mammalian brain tissue using X-ray absorption spectroscopy. We have located and identified individual iron anomalies in an avian tissue model associated with ferritin, biogenic magnetite and haemoglobin with a pixel resolution of less than 5 mu m. This technique represents a breakthrough in the study of both intra- and extra-cellular iron compounds in brain tissue. The potential for high-resolution iron mapping using microfocused X-ray beams has direct application to investigations of the location and structural form of iron compounds associated with human neurodegenerative disorders - a problem which has vexed researchers for 50 years.

Michno K, Van De Hoef D, Wu H, Boulianne GL. 2005. Demented flies? using Drosophila to model human neurodegenerative diseases. Clin Genet 67(6): 468-475.
Abstract: The success of biomedical research in the past few decades has led to dramatic improvements in human health and, as a result, increased life expectancy. An unexpected consequence, however, has been an increase in the number of age-related diseases and, in particular, neurodegenerative diseases. Despite their prevalence, a therapeutic void exists in part due to an incomplete understanding of the biochemical pathogenesis of these diseases. A powerful method that can be used to understand the basic mechanisms underlying neurodegenerative diseases is to generate animal models based on manipulating the expression of single genes that are disease causative. This approach has been facilitated by the fact that many neurodegenerative diseases are inherited as autosomal dominant traits such that expression of the mutant gene in a model organism might be expected to recapitulate the disease. During the past few years, the fruit fly, Drosophila melanogaster, has emerged as a powerful tool to model human neurodegenerative diseases. Here, we describe the various approaches utilized to create fly models of human neurodegenerative disease, and how they can aid in our understanding of disease pathogenesis and facilitate drug discovery and testing.

Mehhase J, Sandig G, Pantopoulos K, Grune T. 2005. Oxidation-induced ferritin turnover in microglial cells: role of proteasome. Free Radic Biol Med 38(2): 276-285.
Abstract: Highly oxidized protein aggregates accumulating in the brain

during neurodegenerative diseases are often surrounded by microglia. Most of the microglial cells surrounding these plaques are activated and release a high amount of oxidizing species. In order to develop their toxic effects numerous oxidizing species need iron. To prevent this iron-dependent oxidation an iron-sequestering apparatus exists, including the major iron storage protein ferritin. Microglial cells damage their own protein pool during activation and it is still unknown whether microglial cells are able to maintain their iron-dependent function during oxidative stress. Therefore, we explored the microglial cell line RAW to test the maintenance of ferritin under oxidizing conditions. Our investigations revealed a half-life of both ferritin chains of 3-3.5 h and a reduced half-life due to oxidation. This was due to the removal of oxidized ferritin by the proteasomal system. Ferritin de novo synthesis was also severely affected by oxidation. This results in a decreased ferritin pool due to acute oxidative stress. These data let us conclude that microglial cells do not increase their ferritin amount after oxidative stress and an increase in the iron storage capacity in these cells after treatment might be achieved only by a high iron saturation of the existing ferritin molecules. (C) 2004 Elsevier Inc. All rights reserved.

Mcfarlane D, Cribb AE. 2005. Systemic and pituitary pars intermedia antioxidant capacity associated with pars intermedia oxidative stress and dysfunction in horses. Am J Vet Res 66(12):2065-2072.
Abstract: Objective-To determine whether a deficiency in systemic or local (pars intermedia) antioxicant capacity is associated with pituitary pars intermedia oxidative stress and pituitary pars intermedia dysfunction (PPID) in horses. Sample Population-Blood samples from 20 horses with PPID and 20 healthy client-owned horses, archived paraffin-em bedded adrenal gland and substantia nigra tissues from 20 horses, and pituitary gland tissue from 16 horses. Procedures-Total glutathione, superoxide dismutase, and glutathione peroxicase activities were determined in RBCs. Accumulation of a systemic marker of oxidative stress (3-nitrotyrosine) was assessed in plasma and formalin-fixed, paraffin-embedded adrenal gland and substantia nigra tissues. Local antioxicants (total and manganese superoxide dismutase, glutathione peroxicase, and total glutathione) were measured in pars intermedia tissues. Results-No significant differences existed in systemic antioxidant enzyme activity or accumulation of 3-nitrotyrosine between horses with PPID and control horses. In pituitary gland tissues, glutathione peroxicase activity was increased in horses with oxidative stress, whereas total glutathione concentration and superoxide dismutase activity remained unchanged. There was an age-associated decrease in manganese superoxide dismutase activity in the pars intermedia. Conclusions and Clinical Relevance-There was no evidence of systemic accumulation of oxidative stress markers or deficiencies in antioxidant capacity in horses with PPID, suggesting that these are unlikely to be major predisposing factors in the development of PPID. Manganese superoxide dismutase activity in the pars intermedia decreased significantly with increasing age. Role of an age-associated decrease in antioxicant capacity for the pars intermedia in the development of PPID in horses warrants further investigation.

Martinaud O, Laquerriere A, Guyant-Marechal L, Ahtoy P, Vera P, Sergeant N, Camuzat A, Bourgeois P, Hauw JJ, Campion D, Hannequin D. 2005. Frontotemporal dementia, motor neuron disease and tauopathy: clinical and neuropathological study in a family. Acta Neuropathol (Berl) 110(1): 84-92.
Abstract: We report a familial disorder occurring in three patients that presented as frontotemporal dementia (FTD). A neuropathological study was performed in a 58-year-old patient, who developed FTD 2 years prior to the onset of motor neuron disease (MND), and died at age 62. Lesions indicative of associated MND were observed: neuronal loss in the anterior horns of the spinal cord, Bunina bodies, axonal spheroids, degeneration of the pyramidal tracts, and of FTD: decreased neuronal density and laminar microvacuolation of layers II and III in the frontal and temporal cortex. Ubiquitin-only-immunoreactive changes were found in the spinal cord and

medulla, but were absent from the temporal and frontal cortex. There were also widespread deposits of various neuronal and glial inclusions containing abnormally phosphorylated tau protein, the Western blotting pattern of which was characterized by two major bands of 64 and 69 kDa. There were no abnormalities of the entire coding sequences of microtubule-associated protein tau (MAPT) and copper-zinc superoxide dismutase (SOD1) genes. Our results suggest that FTD associated with MND can be caused by a larger spectrum of neuropathological lesions than commonly accepted.

Marsala M, Hefferan MP, Kakinohana O, Nakamura S, Marsala J, Tomori Z. 2005. Measurement of peripheral muscle resistance in rats with chronic ischemia-induced paraplegia or morphine-induced rigidity using a semi-automated computer-controlled muscle resistance meter. J Neurotrauma 22(11): 1348-1361.
Abstract: In experimental and clinical studies, an objective assessment of peripheral muscle resistance represents one of the key elements in determining the efficacy of therapeutic manipulations (e.g. pharmacological, surgical) aimed to ameliorate clinical signs of spasticity and/or rigidity. In the present study, we characterize a newly developed limb flexion resistance meter which permits a semi-automated, computer-controlled measurement of peripheral muscle resistance (PMR) in the lower extremities during a forced flexion of the ankle in the awake rat. Ischemic paraplegia was induced in Sprague-Dawley rats by transient aortic occlusion (10 min) in combination with systemic hypotension (40 mm Hg). After ischemia the presence of spasticity component was determined by the presence of an exaggerated EMG activity recorded from gastrocnemius muscle after nociceptive or proprioceptive afferent activation and by velocity-dependent increase in muscle resistance. Rigidity was induced by high dose (30 mg/kg, i.p.) of morphine. Animals with defined ischemic spasticity or morphine-induced rigidity were then placed into a plastic restrainer and. a hind paw attached by a tape to a metal plate driven by a computer-controlled stepping motor equipped with a resistance transducer. The resistance of the ankle to rotation was measured under several testing paradigms: (i) variable degree of ankle flexion (40 degrees, 50 degrees, and 60 degrees), (ii) variable speed/rate of ankle flexion (2, 3, and 4 sec), (iii) the effect of inhalation anesthesia, (iv) the effect of intrathecal baclofen, (v) the effect of dorsal L2-L5 rhizotomy, or (vi) systemic naloxone treatment. In animals with ischemic paraplegia an increased EMG response after peripheral nociceptive or proprioceptive activation was measured. In control animals average muscle resistance was 78 mN and was significantly increased in animals with ischemic spasticity (981-7900 mN). In ischemic-spastic animals a significant increase in measured muscle resistance was seen after increased velocity (4 > 3 > 2 sec) and the angle (40 degrees > 50 degrees > 60 degrees) of the ankle rotation. In spastic animals, deep halothane anesthesia, intrathecal baclofen or dorsal rhizotomy decreased muscle resistance to 39-80% of pretreatment values. Systemic treatment with morphine induced muscle rigidity and corresponding increase in muscle resistance. Morphine-induced increase in muscle resistance was independent on the velocity of the ankle rotation and was reversed by naloxone. These data show that by using this system it is possible to objectively measure the degree of peripheral muscle resistance. The use of this system may represent a simple and effective experimental tool in screening new pharmacological compounds and/or surgical manipulations targeted to modulate spasticity and/or rigidity after a variety of neurological disorders such as spinal cord traumatic or ischemic injury, multiple sclerosis, cerebral palsy, or Parkinson's disease.

Mandel SA, Avramovich-Tirosh Y, Reznichenko L, Zheng HL, Weinreb O, Amit T, Youdim MBH. 2005. Multifunctional activities of green tea catechins in neuroprotection - Modulation of cell survival genes, iron-dependent oxidative stress and PKC signaling pathway. Neurosignals 14(1-2):46-60.
Abstract: Many lines of evidence suggest that oxidative stress resulting in reactive oxygen species (ROS) generation and inflammation play a pivotal role in the age-associated cognitive decline and neuronal loss in neurodegenerative diseases including Alzheimer's (AD), Parkinson's (PD)

and Huntington's diseases. One cardinal chemical pathology observed in these disorders is the accumulation of iron at sites where the neurons die. The buildup of an iron gradient in conjunction with ROS (superoxide, hydroxyl radical and nitric oxide) are thought to constitute a major trigger in neuronal toxicity and demise in all these diseases. Thus, promising future treatment of neurodegenerative diseases and aging depends on availability of effective brain permeable, iron-chelatable/radical scavenger neuroprotective drugs that would prevent the progression of neurodegeneration. Tea flavonoids (catechins) have been reported to possess potent iron-chelating, radical-scavenging and anti-inflammatory activities and to protect neuronal death in a wide array of cellular and animal models of neurological diseases. Recent studies have indicated that in addition to the known antioxidant activity of catechins, other mechanisms such as modulation of signal transduction pathways, cell survival/death genes and mitochondrial function, contribute significantly to the induction of cell viability. This review will focus on the multifunctional properties of green tea and its major component (-)-epigallocatechin-3-gallate (EGCG) and their ability to induce neuroprotection and neurorescue in vitro and in vivo. In particular, their transitional metal (iron and copper) chelating property and inhibition of oxidative stress. Copyright (C) 2005 S. Karger AG, Basel.

Mandel S, Weinreb O, Amit T, Youdim MBH. 2005. Mechanism of neuroprotective action of the anti-Parkinson drug rasagiline and its derivatives. Brain Research Reviews 48(2. Sp. Iss. Si):379-387.
Abstract: The mitochondria are directly involved in cell survival and death. Drugs that protect mitochondria viability and prevent apoptotic cascade mechanisms involved in mitochondrial permeability transition pore (MPTp) will be cytoprotective. Rasagiline (N-propargyl-1R-aminoindan) is a novel, highly potent irreversible monoamine oxidase (MAO) B inhibitor, anti-Parkinson drug. Unlike selegiline, rasagiline is not derived from amphetamine, is not metabolized to neurotoxic I-methamphetamine derivative, nor does it have sympathomimetic activity. Rasagiline is effective as monotherapy or adjunct to L-dopa for patients with early and late Parkinson's disease (PD), and adverse events do not occur with greater frequency in subjects receiving rasagiline than those on placebo. Controlled studies indicate that it might have a disease-modifying effect in PD that may be related to neuroprotection. Its S-isomer, TVP 1022, is a relatively inactive MAO inhibitor. However, both drugs have similar neuroprotective activities in neuronal cell cultures in response to various neurotoxins and in vivo (global ischemia, neurotrauma, head injury, anoxia, etc.), indicating that MAO inhibition is not a pre-requisite for neuroprotection. Structure activity studies have shown that the neuroprotective activity is associated with the propargyl moiety of rasagiline, which protects mitochondrial viability and MPTp by activating Bel-2 and protein kinase C (PKC), and down regulating pro-apoptotic FAS and Bax. Rasagiline and its derivatives also process amyloid precursor protein (APP) to the neuroprotective-neurotrophic soluble APP alpha (sAPP alpha) by PKC and MAP kinase-dependent activation of alpha-secretase. The neuroprotective activity of propargylamine has led us to develop novel bifunctional neuroprotective iron-chelating MAO-inhibiting drugs possessing propargyl moiety for the treatment of other neurodegenerative diseases. (c) 2005 Elsevier B.V. All rights reserved.

Malecki EA, Reich SG, Moliterno AR, Corse AM, Lee LA, Vogelsang GB. 2005. Manganese-induced Parkinsonism from total parenteral nutrition: report of a case and review of the literature. J Neurochem 94:100.

Maharaj H, Maharaj DS, Scheepers M, Mokokong R, Daya S. 2005. L-DOPA administration enhances 6-hydroxydopamine generation. Brain Res 1063 (2):180-186.
Abstract: The therapeutic success of L-3,4-dihydroxyphenylalanine (L-DOPA) treatment in Parkinson's disease (PD) patients remains controversial as many patients become tolerant requiring higher dosage regimens. However, the increase in dosage regimens results in the patients

experiencing intolerable side effects. This study sought to investigate whether dopamine (DA) can chemically react with iron to form the potent neurotoxin 6-hydroxydopamine (6-OHDA). Furthermore, rats were treated with L-DOPA for a period of 7 and 28 days to determine whether L-DOPA treatment results in 6-OHDA formation in rat striatum. In addition, this study also investigates the complex interactions of L-DOPA with iron by performing in vitro and in vivo lipid peroxidation studies and the detection of endogenous 6-OHDA in iron-infused rats. In each study, melatonin was used to determine whether it could quench any free radical effects that may occur. The results of the present study show that DA chemically reacts with iron to form 6-OHDA. Moreover, L-DOPA treatment results in endogenous 6-OHDA formation in rat brain as well as enhances iron-induced lipid peroxidation both in vitro and in vivo in the rat striatum. The L-DOPA-induced increase in lipid peroxidation, in iron-infused rats, corresponds with an increase in levels of 6-OHDA in the rat striatum. The use of melatonin significantly decreases the L-DOPA-stimulated 6-OHDA formation in the rat striatum. The present study provides novel information on L-DOPA-induced neurotoxicity and suggests the concomitant use of an antioxidant with L-DOPA in order to enhance the life span of L-DOPA therapy. (C) 2005 Elsevier B.V. All rights reserved.

Lu L, Zhang LI, Li GJ, Guo WR, Liang WN, Zheng W. 2005. Alteration of serum concentrations of manganese, iron, ferritin, and transferrin receptor following exposure to welding fumes among career welders. Neurotoxicology 26(2):257-265.
Abstract: This study was performed to determine airborne manganese levels during welding practice and to establish the relationship between long-term, low-level exposure to manganese and altered serum concentrations of manganese, iron, and proteins associated with iron metabolism in career welders. Ninety-seven welders (average age of 36 years) who have engaged in electric arc weld in a vehicle manufacturer were recruited as the exposed group. Welders worked 7-8 h per day with employment duration of 1-33 years. Control subjects consisted of 91 employees (average age of 35 years) in the same factory but not in the welding profession. Ambient manganese levels in welders' breathing zone were the highest inside the vehicle (1.5 +/- 0.7 mg/m(3)), and the lowest in the center of the workshop (0.2 +/- 0.05 mg/m(3)). Since the filter size was 0.8 mu m, it is possible that these values may be likely an underestimation of the true manganese levels. Serum levels of manganese and iron in welders were about three-fold ($p < 0.01$) and 1.2-fold ($p < 0.01$), respectively, higher than those of controls. Serum concentrations of ferritin and transferrin were increased among welders, while serum transferrin receptor levels were significantly decreased in comparison to controls. Linear regression analyses revealed a lack of association between serum levels of manganese and iron. However serum concentrations of iron and ferritin were positively associated with Years of welder experience ($p < 0.05$). Moreover, serum transferrin receptor levels were inversely associated with serum manganese concentrations ($p < 0.05$). These findings suggest that exposure to welding fume among welders disturbs serum homeostasis of manganese, iron, and the proteins associated with iron metabolism. Serum manganese may serve as a reasonable biomarker for assessment of recent exposure to airborne manganese. (C) 2004 Elsevier Inc. All rights reserved.

Lotharius J, Falsig J, Van Beek J, Payne S, Dringen R, Brundin P, Leist M. 2005. Progressive degeneration of human mesencephalic neuron-derived cells triggered by dopamine-dependent oxidative stress is dependent on the mixed-lineage kinase pathway. J Neurosci 25(27):6329-6342.
Abstract: Models of Parkinson's disease (PD) based on selective neuronal death have been used to study pathogenic mechanisms underlying nigral cell death and in some instances to develop symptomatic therapies. For validation of putative neuroprotectants, a model is desirable in which the events leading to neurodegeneration replicate those occurring in the disease. We developed a human in vitro model of PD based on the assumption that dysregulated cytoplasmic dopamine levels trigger cell loss

in this disorder. Differentiated human mesencephalic neuron-derived cells were exposed to methamphetamine (METH) to promote cytoplasmic dopamine accumulation. In the presence of elevated iron concentrations, as observed in PD, increased cytosolic dopamine led to oxidative stress, c-Jun N-terminal kinase (JNK) pathway activation, neurite degeneration, and eventually apoptosis. We examined the role of the mixed-lineage kinases (MLKs) in this complex degenerative cascade by using the potent inhibitor 3,9-bis[(ethylthio) methyl]-K-252a (CEP1347). Inhibition of MLKs not only prevented FeCl2+/METH- induced JNK activation and apoptosis but also early events such as neurite degeneration and oxidative stress. This broad neuroprotective action of CEP1347 was associated with increased expression of an oxidative stress-response modulator, activating transcription factor 4. As a functional consequence, transcription of the cystine/glutamate and glycine transporters, cellular cystine uptake and intracellular levels of the redox buffer glutathione were augmented. In conclusion, this new human model of parkinsonian neurodegeneration has the potential to yield new insights into neurorestorative therapeutics and suggests that enhancement of cytoprotective mechanisms, in addition to blockade of apoptosis, may be essential for disease modulation.

Logroscino G, Chen HL, Wing A, Ascherio A. 2005. Blood donations, iron stores and risk of Parkinson's disease. Neurology 64(6):A311 .

Liu LL, Franz KJ. 2005. Phosphorylation of an alpha-synuclein peptide fragment enhances metal binding. J Am Chem Soc 127(27):9662-9663.

Liu G, Garrett MR, Men P, Zhu XW, Perry G, Smith MA. 2005. Nanoparticle and other metal chelation therapeutics in Alzheimer disease. Biochimica Et Biophysica Acta-Molecular Basis of Disease 1741(3):246-252.
Abstract: Current therapies for Alzheimer disease (AD) such as the anticholinesterase inhibitors and the latest NMDA receptor inhibitor, Namenda, provide moderate symptomatic delay at various stages of disease, but do not arrest disease progression or supply meaningful remission. As such, new approaches to disease management are urgently needed. Although the etiology of AD is largely unknown, oxidative damage mediated by metals is likely a significant contributor since metals such as iron, aluminum, zinc, and copper are dysregulated and/or increased in AD brain tissue and create a pro-oxidative environment. This role of metal ion-induced free radical formation in AD makes chelation therapy an attractive means of dampening the oxidative stress burden in neurons. The chelator desferioxamine, FDA approved for iron overload, has shown some benefit in AD, but like many chelators, it has a host of adverse effects and substantial obstacles for tissue-specific targeting. Other chelators are under development and have shown various strengths and weaknesses. In this review, we propose a novel system of chelation therapy through the use of nanoparticles. Nanoparticles conjugated to chelators show a unique ability to cross the blood-brain barrier (131313), chelate metals, and exit through the BBB with their corresponding complexed metal ions. This method may prove to be a safe and effective means of reducing the metal load in neural tissue thus staving off the harmful effects of oxidative damage and its sequelae. (c) 2005 Elsevier B.V. All rights reserved.

Li J, Scheller C, Koutsilieri E, Griffiths F, Beart PM, Mercer LD, Halliday G, Kettle E, Rowe D, Riederer P, Gerlach M, Rodriguez M, Double KL. 2005. Differential effects of human neuromelanin and synthetic dopamine melanin on neuronal and glial cells. J Neurochem 95(2):599-608.
Abstract: We investigated the effects of neuromelanin (NM) isolated from the human substantia nigra and synthetic dopamine melanin (DAM) on neuronal and glial cell lines and on primary rat mesencephalic cultures. Lactate dehydrogenase (LDH) activity and lipid peroxidation were significantly increased in SK-N-SH cells by DAM but not by NM. In contrast, iron-saturated NM significantly increased LDH activity in SK-N-SH cells, compared with 100 mg/mL ETDA-treated NM containing a low concentration of bound iron. DAM, but not NM, stimulated hydroxyl radical production and increased SK-N-SH cell death via apoptotic-like mechanisms. Neither DAM

nor NM induced any changes in the glial cell line U373. H-3-Dopamine uptake in primary rat mesencephalic cultures was significantly reduced in DAM- compared with NM-treated cultures, accompanied by increased cell death via an apoptosis-like mechanism. Interestingly, Fenton-induced cell death was significantly decreased in cultures treated with both Fenton reagent and NM, an effect not seen in cultures treated with Fenton reagent plus DAM. These data are suggestive of a protective role for neuromelanin under conditions of high oxidative load. Our findings provide new evidence for a physiological role for neuromelanin in vivo and highlights the caution with which data based upon model systems should be interpreted.

Li GJ, Zhao QQ, Zheng W. 2005. Alteration at translational but not transcriptional level of transferrin receptor expression following manganese exposure at the blood-CSF barrier in vitro. Toxicol Appl Pharmacol 205(2):188-200.
Abstract: Manganese exposure alters iron homeostasis in blood and cerebrospinal fluid (CSF), possibly by acting on iron transport mechanisms localized at the blood-brain barrier and/or blood-CSF barrier. This study was designed to test the hypothesis that manganese exposure may change the binding affinity of iron regulatory proteins (IRPs) to rnRNAs encoding transferrin receptor (TfR), thereby influencing iron transport at the blood-CSF barrier. A primary culture of choroidal epithelial cells was adapted to grow on a permeable membrane sandwiched between two culture chambers to mimic blood-CSF barrier. Trace Fe-59 was used to determine the transepithelial transport of iron. Following manganese treatment (100 mu M for 24 h), the initial flux rate constant (K-i) of iron was increased by 34%, whereas the storage of iron in cells was reduced by 58%, as compared to controls. A gel shift assay demonstrated that manganese exposure increased the binding of IRP1 and IRP2 to the stein loop-containing mRNAs. Consequently, the cellular concentrations of TfR proteins were increased by 84% in comparison to controls. Assays utilizing RT-PCR, quantitative real-time reverse traiiscriptase-PCR, and nuclear run off techniques showed that manganese treatment did not affect the level of heterogeneous nuclear RNA (hnRNA) encoding TfR, nor did it affect the level of nascent TfR mRNA. However, manganese exposure resulted in a significantly increased level of TfR mRNA and reduced levels of ferritin mRNA. Taken together, these results suggest that manganese exposure increases iron transport at the blood-CSF barrier; the effect is likely due to manganese action on translational events relevant to the production of TfR, but not due to its action on transcriptional, gene expression of TfR. The disrupted protein-TfR rnRNA interaction in the choroidal epithelial cells may explain the toxicity of manganese at the blood-CSF barrier. (c) 2004 Elsevier Inc. All rights reserved.

Lewis J, Bench G, Myers O, Tinner B, Staines W, Barr E, Divine KK, Barrington W, Karlsson J. 2005. Trigeminal uptake and clearance of inhaled manganese chloride in rats and mice. Neurotoxicology 26(1):113-123.
Abstract: Inhaled manganese (Mn) can enter the olfactory bulbs via the olfactory epithelium, and can then be further transported trans-synaptically to deeper brain structures. In addition to olfactory neurons, the nasal cavity is innervated by the maxillary division of the trigeminal nerve that projects to the spinal trigerninal nucleus. Direct uptake and transport of inhaled metal particles in the trigeminal system has not been investigated previously. We studied the uptake, deposition, and clearance of soluble Mn in the trigeminal system following nose-only inhalation of environmentally relevant concentrations. Rats and mice were exposed for 10-days (6 h/day, 5 days/week) to air or MnCl2 aerosols 3 ;containing 2.3 +/- 1.3 mg/m (3) Mn with mass median aerodynamic diameter (MMAD) of 3.1 +/- 1.4 mum for rats and 2. 0 +/- 0.09 mg/n(3) Mn MnCl2 with MMAD of 1.98 +/- 0.12 mum for mice. Mn concentrations in the trigeminal ganglia and spinal trigeminal nucleus were measured 2 h (0-day), T, 14-, or 30-days post-exposure using proton induced X-ray emission (PIXE). Manganese-exposed rats and mice showed statistically elevated levels of Mn in trigeminal ganglia 0-, 7- and 14days after the 10-days exposure period when compared to control animals. The Mn concentration gradually decreased over time with a clearance rate (t(1/2)) of 7-8-days. Rats and mice were

similar in both average accumulated Mn levels in trigeminal ganglia and in rates of clearance. We also found a small but significant elevation of Mn in the spinal trigeminal nucleus of mice 7-days post-exposure and in rats 0- and 7-days post-exposure. Our data demonstrate that the trigeminal nerve can serve as a pathway for entry of inhaled Mn to the brain in rodents following nose-only exposure and raise the question of whether entry of toxicants via this pathway may contribute to development of neurodegenerative diseases. (C) 2004 Elsevier Inc. All rights reserved.

Levin OS. 2005. "Ephedron" encephalopathy. Zh Nevrol Psikhiatr Im S S Korsakova 105(7):12-20.
Abstract: The results of clinical, neuropsychological and MRI study of 21 patients with "ephedron" encephalopathy caused by intake of methcatinon ("ephedron"), a surrogate drug obtained from phenylpropanolamine-containing compounds by adding potassium permanganate, are presented. Signs of brain lesions emerged 3-14 (mean 6,8 +/- 4,9) months after the beginning of the regular drug intake. Neurological disturbances were measured using the Scale of clinical assessment of ephedron encephalopathy. In the acute stage of the disease, most patients had the combination of extrapyramidal disorders (parkinsonism, muscular dystonia, tremor, myoclonia) with pronounced postural instability, pseudobulbar syndrome, autonomic, cognitive and affective personality abnormalities of subcortical and frontal types. In 18 (86%) patients, MRI revealed a bilateral symmetric elevation of the signal from the basal ganglia on T1-weighted images, mostly from the medial segment of globus pallidus and the reticular part of substantia nigra that reflected magnesium accumulation. The spread of hyperintensive MRI changes negatively correlated with the disease duration (r=-0,6; p<0,01), but did not depend on the drug abuse duration or its approximate total dosage, and also did not correspond to the disease severity. In follow-up, a tendency to spontaneous regress of symptoms was observed in 29% of the cases, and in 33% patients symptoms have been regressing even 4 years after stopping of methcatinon intake. The main mechanisms of "ephedron" encephalopathy development are probably related to the manganese accumulation in the brain that might trigger secondary pathogenetic mechanisms, such as mitochondrial dysfunction, oxidative stress, etc. The induction courses of calcium and sodium EDTA that accelerates manganese excretion decrease a probability of the further disease progress, though do not contribute significantly to symptoms regress. The data on possibilities of symptomatic therapy of movement and affective disturbances is presented.

Levi S, Cozzi A, Arosio P. 2005. Neuroferritinopathy: a neurodegenerative disorder associated with L-ferritin mutation. Best Practice & Research Clinical Haematology 18(2):265-276.
Abstract: Neuroferritinopathy is a dominantly inherited movement disorder characterized by deposition of iron and ferritin in the brain, normal or low serum ferritin levels, and highly variable clinical features. The disease, also named dominant adult-onset basal ganglia disease, is associated with a nucleotide insertion that modifies the last 22 amino acids of the ferritin L-chain. A similar dominant movement disorder in a French family was associated with a nucleotide insertion that modifies the last nine amino acids of the same molecule. Both disorders show ferritin and iron precipitates in the basal ganglia of the brain. Here we present the structural aspects of the two mutations, as well studies on cellular models aimed at understanding the molecular basis of the disorder. The results indicate that the mutations affect protein folding and stability, and that the expression of one of the two variant ferritins increases intracellular iron availability and sensitivity to oxidative damage.

Levenson CW. 2005. Trace metal regulation of neuronal apoptosis: From genes to behavior. Physiology & Behavior 86(3):399-406.
Abstract: The genetically programmed form of neuronal death known as apoptosis plays a role in many neurodegenerative diseases including Alzheimer's disease, Parkinson's disease, amyotrophic lateral sclerosis

(ALS) and Huntington's disease. Apoptosis is also responsible for neuronal death after traumatic brain and spinal cord injury, stroke, and seizures. The cognitive and behavioral consequences of all of these disorders can be devastating. Unfortunately the mechanisms that regulate neuronal apoptosis are complex. However, it is this very complexity that provides us with a wide array of potential targets for the development of anti-apoptotic strategies. Thus, our lab is currently exploring the molecular and cellular mechanisms responsible for neuronal apoptosis, with a particular focus on the role of the metals copper, zinc, and iron. Each of these metals is essential for normal central nervous system (CNS) development and function. However, imbalances, either excess or deficiency. can result in neuronal apoptosis. In this review, we show the relationship between these metals in neurodegenerative disorders and CNS injury, and the mechanisms that govern neuronal survival and apoptosis. (c) 2005 Elsevier Inc. All rights reserved.

Latchoumycandane C, Anantharam V, Kitazawa M, Yang YJ, Kanthasamy A, Kanthasamy AG. 2005. Protein kinase C delta is a key downstream mediator of manganese-induced apoptosis in dopaminergic neuronal cells. J Pharmacol Exp Ther 313(1): 46-55.
Abstract: Manganese (Mn) exposure causes manganism, a neurological disorder similar to Parkinson's disease. However, the cellular mechanism by which Mn induces dopaminergic neuronal cell death remains unclear. In the present study, we sought to investigate the key downstream apoptotic cell signaling events that contribute to Mn-induced cell death in mesencephalic dopaminergic neuronal (N27) cells. Mn exposure induced a dose-dependent increase in neuronal cell death in N27 cells. The cell death was accompanied by sequential activation of mitochondrial-dependent proapoptotic events, including cytochrome c release, caspase-3 activation, and DNA fragmentation, but not caspase-8 activation, indicating that the mitochondrial-dependent apoptotic cascade primarily triggers Mn-induced apoptosis. Notably, Mn treatment proteolytically activated protein kinase C delta (PKC delta), a member of a novel class of protein kinase C. The caspase-3 specific inhibitor benzyloxycarbonyl-Asp-Glu-Val-Asp-fluoromethylketone (Z-DEVD-FMK) significantly blocked PKC delta cleavage and its kinase activity, indicating that caspase-3 mediates the proteolytic activation. Cotreatment with the PKC delta inhibitor rottlerin or the caspase-3 inhibitor Z-DEVD-FMK almost completely blocked Mn-induced DNA fragmentation. Additionally, N27 cells expressing a catalytically inactive PKC delta(K376R) protein (PKC delta dominant negative mutant) or a caspase cleavage resistant PKC delta(D327A) protein (PKC delta cleavage resistant mutant) were found to be resistant to Mn-induced apoptosis. To further establish the proapoptotic role of PKC delta, RNA interference-mediated gene knockdown was performed. Small interfering RNA suppression of PKC delta expression protected N27 cells from Mn-induced apoptotic cell death. Collectively, these results suggest that caspase-3-dependent proteolytic activation of PKC delta plays a key role in Mn-induced apoptotic cell death.

Landrigan PJ, Sonawane B, Butler RN, Trasande L, Callan R, Droller D. 2005. Early environmental origins of neurodegenerative disease in later life. Environ Health Perspect 113(9):1230-1233.
Abstract: Parkinson disease (PD) and Alzheimer disease (AD), the two most common neurodegenerative disorders in American adults, are of purely genetic origin in a minority of cases and appear in most instances to arise through interactions among genetic and environmental factors. In this article we hypothesize that environmental exposures in early life may be of particular etiologic importance and review evidence for the early environmental origins of neurodegeneration. For PD the first recognized environmental cause, MPTP (1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine), was identified in epidemiologic studies of drug abusers. Chemicals experimentally linked to PD include the insecticide rotenone and the herbicides paraquat and maneb; interaction has been observed between paraquat and maneb. In epidemiologic studies, manganese has been linked to parkinsonism. In dementia, lead is

associated with increased risk in chronically exposed workers. Exposures of children in early life to lead, polychlorinated biphenyls, and methylmercury have been followed by persistent decrements in intelligence that may presage dementia. To discover new environmental causes of AD and PD, and to characterize relevant gene-environment interactions, we recommend that a large, prospective genetic and epidemiologic study be undertaken that will follow thousands of children from conception (or before) to old age. Additional approaches to etiologic discovery include establishing incidence registries for AD and PD, conducting targeted investigations in high-risk populations, and improving testing of the potential neurologic toxicity of chemicals.

Kowalik-Jankowska T, Rajewska A, Wisniewska K, Grzonka Z, Jezierska J. 2005. Coordination abilities of N-terminal fragments of alpha-synuclein towards copper(II) ions: A combined potentiometric and spectroscopic study. J Inorg Biochem 99(12):2282-2291.
Abstract: Copper(II) complexes of the 1-17 (MDVFMKGLSKAKEGVVA-NH2), 1-28 (MDVFMKGLSKAKEGVVAAAEKTKQGVAENHz), 1-39 (MDVFMKGLSKAKEGVVAAAEKTKQGVAEAPGKTKEGVLY-NH2) and 1-39 (A30P) fragments of a-synuclein were studied by potentiometric, UV-Vis (UV-visible), CD (circular dichroism) and EPR (electron paramagnetic resonance) spectroscopic methods to determine the stoichiometry, stability constants and coordination modes of the complexes formed. The (beta-carboxylate group of Asp residue in second position of the peptide chain coordinates strongly to Cu(II) ion over the pH range 4-9.5 to give unusually stable 2N complex with (NH2, N-; beta-COO-. H2O) coordination mode. At pH above 7 the results suggest the formation of 2N, 3N, 4N complexes (in equatorial plane) and the involvement of the lateral NH2 group of Lys residue in the axial coordination of Cu(II) ion. In CD spectra sigma (epsilon-NH2-Lys) -> Cu(II) charge transfer transition is observed. Addition of the 18-28 and 18-39 fragments to the 1-17 peptide does not change the coordination mode and the 1-39 fragment forms the Cu(II) complexes with higher stabilities compared to those of the 1-17, 1-28 and 1-39(A30P) fragments of x-synuclein. (c) 2005 Elsevier Inc. All rights reserved.

Kotzbauer PT, Truax AC, Trojanowski JQ, Lee VMY. 2005. Altered neuronal mitochondrial coenzyme a synthesis in neurodegeneration with brain iron accumulation caused by abnormal processing, stability, and catalytic activity of mutant pantothenate kinase 2. J Neurosci 25(3):689-698.
Abstract: Mutations in the pantothenate kinase 2 (PANK2) gene have been identified in patients with neurodegeneration with brain iron accumulation (NBIA; formerly Hallervorden - Spatz disease). However, the mechanisms by which these mutations cause neurodegeneration are unclear, especially given the existence of multiple pantothenate kinase genes in humans and multiple PanK2 transcripts with potentially different subcellular localizations. We demonstrate that PanK2 protein is localized to mitochondria of neurons in human brain, distinguishing it from other pantothenate kinases that do not possess mitochondrial-targeting sequences. PanK2 protein translated from the most 5 5' start site is sequentially cleaved at two sites by the mitochondrial processing peptidase, generating a long-lived 48 kDa mature protein identical to that found in human brain extracts. The mature protein catalyzes the initial step in coenzyme A (CoA) synthesis but displays feedback inhibition in response to species of acyl CoA rather than CoA itself. Some, but not all disease-associated point mutations result in significantly reduced catalytic activity. The most common mutation, G521R, results in marked instability of the intermediate PanK2 isoform and reduced production of the mature isoform. These results suggest that NBIA is caused by altered neuronal mitochondrial lipid metabolism caused by mutations disrupting PanK2 protein levels and catalytic activity.

Kotamraju S, Kalivendi S, Shang T, Kalyanaraman B. 2005. Nitric Oxide, Proteasomal Function, and Iron Homeostasis - Implications in Aging and Neurodegenerative DiseasesVolume 396. p 526-+. Nitric Oxide, Pt E:

Methods in Enzymology.
Abstract: In this chapter, oxidant-induced transferrin receptor-mediated iron-signaling and apoptosis are described in endothelial and neuronal cells exposed to oxidants. The role of nitric oxide in the regulation of iron homeostasis and oxidant-induced apoptosis is described. The interrelationship between oxidative stress, iron-signaling, and nitric oxide-dependent proteasomal function provides a rational mechanism that connects both oxidative and nitrative modifications.

Koike Y, Frey MA, Sahiar F, Dodge R, Mohler S. 2005. Effects of HZE particle on the nigrostriatal dopaminergic system in a future mars mission. Acta Astronautica 56(3):367-378.
Abstract: Because of long duration travel outside the Earth's magnetic field, the effect of iron-rich high charge and energy (HZE) particles in Galactic Cosmic Rays on human body is the major concern in radiation protection. Recently attention has been directed to effects on the central nervous system in addition to mutagenic effects. In particular, a reduction in striatal dopamine content on nigrostriatal dopaminergic system has been reported by investigators using accelerated iron ions in ground-based mammalian studies. In addition, studies of the pathophysiology of Parkinson's disease demonstrated that excess iron cause a reduction in the dopamine content in the substantia nigra. This suggests an intriguing possibility to explain the selective detrimental effects of HZE particles on the dopaminergic system. Should these particles have biochemical effects, possible options for countermeasures are: (1) nutritional prevention, (2) medication, and (3) surgical placement of a stimulator electrode at a specific anatomic site in the basal ganglia. (C) 2004 Elsevier Ltd. All rights reserved.

Klos KJ, Ahlskog JE, Josephs DA, Fealey RD, Cowl CT, Kumar N. 2005. Neurologic spectrum of chronic liver failure and basal ganglia T1 hyperintensity on magnetic resonance imaging - Probable manganese neurotoxicity. Arch Neurol 62(9):1385-1390.
Abstract: Background: An atypical form of parkinsonism has been described in patients with chronic liver disease, associated with increased T1 signal in the basal ganglia on magnetic resonance imaging. The magnetic resonance imaging signal changes are characteristic of manganese accumulation, which has been neuropathologically confirmed. Manganese neurotoxicity may result in additional neurologic findings besides parkinsonism. Objective: To fully characterize patients with chronic central nervous system symptoms and chronic liver failure associated with basal ganglia T1 hyperintensity. Design: Prospective and retrospective case study Setting: Mayo Clinic, Rochester, Minn. Participants: Eight Patients referred for neurologic evaluation and studied prospectively, and 7 additional retrospectively identified patients who had been examined by Mayo Clinic neurologists. Main Outcome Measures: Neurologic syndromes identified. Results: Three syndromes were recognized in these 15 patients with liver failure and basal ganglia T1 hyperintensity on magnetic resonance imaging: (1) isolated parkinsonism, (2) gait ataxia plus other neurologic findings (ataxia-plus), and (3) cognitive impairment with psychiatric features. All but I patient had elevated blood manganese levels. Ammonia levels were normal in most, and the neurologic syndromes did not appear to reflect the well-known toxic-metabolic encephalopathy of liver disease. Conclusions: Chronic liver failure may result in heterogeneous neurologic syndromes that cut across a variety of liver diseases. We selected cases on the basis of evidence of brain manganese accumulation, and this may be a crucial component of these syndromes. Further studies are necessary to explore this issue.

Klopstock T, Elstner M, Lucking CB, Muller-Myhsok B, Gasser T, Botz E, Lichtner P, Hortnagel K. 2005. Mutations in the pantothenate kinase gene PANK2 are not associated with Parkinson disease. Neurosci Lett 379(3):195-198.
Abstract: Pantothenate kinase-associated neurodegeneration (PKAN) may serve as a model for Parkinson disease (PD) since many PKAN patients suffer from parkinsonism and both conditions lead to iron accumulation in

the basal ganglia. We screened the gene coding for pantothenate kinase 2 (PANK2) for sequence variants in PD. We found no mutations in 67 PD patients with affected sibs or early-onset disease. Moreover, PANK2 polymorphisms were not associated with late-onset idiopathic PD in 339 patients. We conclude that PANK2 variants exert, if any, only a very small effect in the genetic risk of PD. © 2005 Elsevier Ireland Ltd. All rights reserved.

Klein SM, Behrstock S, Mchugh J, Hoffmann K, Wallace K, Suzuki M, Aebischer P, Svendsen CN. 2005. GDNF delivery using human neural progenitor cells in a rat model of ALS. Hum Gene Ther 16(4):509-521.
Abstract: Amyotrophic lateral sclerosis (ALS) is a fatal neurodegenerative disease characterized by progressive loss of spinal cord, brainstem, and cortical motor neurons. In a minority of patients, the disease is caused by mutations in the copper (2+)/zinc (2+) superoxide dismutase 1 (SOD1) gene. Recent evidence suggests that astrocytes are dysfunctional in ALS and may be a critical link in the support of motor neuron health. Furthermore, growth factors, such as glial cell line-derived neurotrophic factor (GDNF), have a high affinity for motor neurons and can prevent their death following various insults, but due to the protein's large size are difficult to directly administer to brain. In this study, human neural progenitor cells (hNPC) isolated from the cortex were expanded in culture and modified using lentivirus to secrete GDNF (hNPC(GDNF)). These cells survived up to 11 weeks following transplantation into the lumbar spinal cord of rats overexpressing the G93A SOD1 mutation (SOD1(G93A)). Cellular integration into both gray and white matter was observed without adverse behavioral effects. All transplants secreted GDNF within the region of cell survival, but not outside this area. Fibers were seen to upregulate cholinergic markers in response to GDNF, indicating it was physiologically active. We conclude that genetically modified hNPC can survive, integrate, and release GDNF in the spinal cord of SOD1(G93A) rats. As such, they provide an interesting source of cells for both glial replacement and trophic factor delivery in future human clinical studies.

Kitzberger R, Madl C, Ferenci P. 2005. Wilson disease. Metab Brain Dis 20(4): 295-302.
Abstract: Wilson disease (WD) is an autosomal recessive inherited disorder of copper metabolism, resulting in pathological accumulation of copper in many organs and tissues. The hallmarks of the disease are the presence of liver disease, neurologic symptoms, and Kayser-Fleischer corneal rings. The leading neurologic symptoms in WD are dysathria, dyspraxia, ataxia, and Parkinsonian-like extrapyramidal signs. Changes in the basal ganglia in brain magnetic resonance imaging (MRI) are characteristic features of the disease. In presence of liver cirrhosis, some features may resemble hepatic encephalopathy. Symptoms and MRI abnormalities may be fully reversible on treatment with zinc or copper chelators. Improvement can be monitored by serial recording of brain-stem-evoked responses. The basic defect is an impaired trafficking of copper in hepatocytes. ATP7B is the gene product of the WD gene located on chromosome 13 and resides in hepatocytes in the trans-Golgi network, transporting copper into the secretory pathway for incorporation into apoceruloplasmin and excretion into the bile. While about 40% of patients preset with neurologic symptoms, little is known about the role of copper and ATP7B in the central nervous system. In some brain areas, like in the pineal gland, ATP7B is expressed and functionally active. Increasing evidence supports an important role for metals in neurobiology. Two proteins related to neurodegeneration are copper-binding proteins (1) the amyloid precursor protein (APP), a protein related to Alzheimer's disease, and (2) the Prion protein, related to Creutzfeldt-Jakob disease. A major source of free-radical production in the brain derives from copper. To prevent metal-mediated oxidative stress, cells have evolved complex metal transport systems. APP is a major regulator of neuronal copper homeostasis and has a copper-binding domain (CuBD). The surface location of this site, structural homology of CuBD to copper chaperones, and the role of APP in neuronal copper homeostasis are consistent with the CuBD acting as a

neuronal metallotransporter. There are several copper-containing enzymes in the brain, like dopamine beta hydroxylase or Cu/Zn superoxide dismutase (SOD1). Their function may be altered because of copper overload. WD appears to be associated with a dopaminergic deficit. Mutations in the SOD1gene cause familial amyotrophic lateral sclerosis. Survival of transgenic mice with a mutant SOD1 which fails to incorporate Cu((2+)) in its active site was improved by copper depletion. Wilson disease (WD) is an autosomal recessive inherited disorder in which copper pathologically accumulates primarily within the liver and subsequently in the neurologic system and many other organs and tissues. Presence of liver disease, neurologic symptoms, and Kayser-Fleischer corneal rings are the hallmarks of the disease.

Kitazawa M, Anantharam V, Yang YJ, Hirata Y, Kanthasamy A, Kanthasamy AG. 2005. Activation of protein kinase C delta by proteolytic cleavage contributes to manganese-induced apoptosis in dopaminergic cells: protective role of Bcl-2. Biochem Pharmacol 69(1):133-146.
Abstract: Chronic inorganic manganese exposure causes selective toxicity to the nigrostriatal dopaminergic system, resulting in a Parkinsonian-like neurological condition known as Manganism. Apoptosis has been shown to occur in manganese-induced neurotoxicity; however, the down-stream cellular target of caspase-3 that contributes to DNA fragmentation is not established. Herein, we demonstrate that proteolytic activation of protein kinase Cdelta (PKCdelta) by caspase-3 plays a critical role in manganese-induced apoptotic cell death. Treatment of PC12 cells with manganese caused a sequential activation of mitochondrial-dependent pro-apoptotic events, including mitochondrial membrane depolarization, cytochrome c release, caspase-3 activation, and DNA fragmentation. Overexpression of Bcl-2 in PC12 cells remarkably attenuated each of these events, indicating that the mitochondrial-dependent apoptotic cascade contributes to manganese-induced apoptosis. Furthermore, PKCdelta was proteolytically cleaved by caspase-3, causing a persistent activation of the kinase. The manganese-induced proteolytic cleavage of PKCdelta was significantly blocked by Bcl-2-overexpression. Administration of active recombinant PKCdelta induced DNA fragmentation in PC12 cells, suggesting a pro-apoptotic role of PKCdelta. Furthermore, expression of catalytically inactive mutant PKCdelta(K376R) via a lentiviral gene delivery system effectively attenuated manganese-induced apoptosis. Together, these results suggest that the mitochondrial-dependent caspase cascade mediates apoptosis via proteolytic activation of PKCdelta in manganese-induced neurotoxicity. (C) 2004 Elsevier Inc. All rights reserved.

Kim YS, Kim SS, Cho JJ, Choi DH, Hwang O, Shin DH, Chun HS, Beal MF, Joh TH. 2005. Matrix metalloproteinase-3: A novel signaling proteinase from apoptotic neuronal cells that activates microglia. J Neurosci 25(14): 3701-3711.
Abstract: Microglial activation and inflammation are associated with progressive neuronal apoptosis in neurodegenerative human brain disorders. We sought to investigate molecular signaling mechanisms that govern activation of microglia in apoptotic neuronal degeneration. We report here that the active form of matrix metalloproteinase-3 (MMP-3) was released into the serum-deprived media (SDM) of PC12 cells and other media of apoptotic neuronal cells within 2-6 h of treatment of the cells, and SDM and catalytic domain of recombinant MMP-3 (cMMP-3) activated microglia in primary microglia cultures as well as BV2 cells, a mouse microglia cell line. Both SDM and cMMP-3 induced generation of tumor necrosis factor alpha(TNF alpha), interleukin-6 (IL-6), IL-1 beta, and interleukin-1 receptor antagonist but not IL-12 and inducible nitric oxide synthase, which are readily induced by lipopolysaccharide, in microglia, suggesting that there is a characteristic pattern of microglial cytokine induction by apoptotic neurons. Neither glial cell line-derived neurotrophic factor nor anti-inflammatory cytokines, such as IL-10 and transforming growth factor-beta 1, were induced. SDM and cMMP-3 extensively released TNF-alpha from microglia and activated the nuclear factor-KB pathway, and these microglial responses were totally abolished by preincubation with an

MMP-3 inhibitor, NNGH [N-isobutyl-N-(4-methoxyphenylsulfonyl)-glycylhydroxamic acid]. MMP-3-mediated microglial activation mostly depended on ERK (extracellular signal-regulated kinase) phosphorylation but not much on either JNK (c-Jun N-terminal protein kinase) or p38 activation. Conditioned medium of SDM-or cMMP-3-activated BV2 cells caused apoptosis of PC12 cells. These results strongly suggest that the distinctive signal of neuronal apoptosis is the release of active form of MMP-3 that activates microglia and subsequently exacerbates neuronal degeneration. Therefore, the release of MMP-3 from apoptotic neurons may play a major role in degenerative human brain disorders, such as Parkinson's disease.

Kim YJ, Nakatomi R, Akagi T, Hashikawa T, Takahashi R. 2005. Unsaturated fatty acids induce cytotoxic aggregate formation of amyotrophic lateral sclerosis-linked superoxide dismutase 1 mutants. J Biol Chem 280(22): 21515-21521.
Abstract: Formation of misfolded protein aggregates is a remarkable hallmark of various neurodegenerative diseases including Alzheimer disease, Parkinson disease, Huntington disease, prion encephalopathies, and amyotrophic lateral sclerosis (ALS). Superoxide dismutase 1 (SOD1) immunoreactive inclusions have been found in the spinal cord of ALS animal models and patients, implicating the close involvement of SOD1 aggregates in ALS pathogenesis. Here we examined the molecular mechanism of aggregate formation of ALS-related SOD1 mutants in vitro. We found that long-chain unsaturated fatty acids (FAs) promoted aggregate formation of SOD1 mutants in both dose- and time-dependent manners. Metal-deficient SOD1s, wild-type, and mutants were highly oligomerized compared with holo-SOD1s by incubation in the presence of unsaturated FAs. Oligomerization of SOD1 is closely associated with its structural instability. Heat-treated holo-SOD1 mutants were readily oligomerized by the addition of unsaturated FAs, whereas wild-type SOD1 was not. The monounsaturated FA, oleic acid, directly bound to SOD1 and was characterized by a solid-phase FA binding assay using oleate-Sepharose. The FA binding characteristics were closely correlated with the oligomerization propensity of SOD1 proteins, which indicates that FA binding may change SOD1 conformation in a way that favors the formation of aggregates. High molecular mass aggregates of SOD1 induced by FAs have a granular morphology and show significant cytotoxicity. These findings suggest that SOD1 mutants gain FA binding abilities based on their structural instability and form cytotoxic granular aggregates.

Kim KS, Kang JH. 2005. Aggregation of alpha-synuclein induced by oxidized catecholamines as a potential mechanism of Lewy body . Bulletin of the Korean Chemical Society 26(8):1255-1259.
Abstract: Lewy bodies (LBs) are neuronal inclusions that are closely related to Parkinson's disease (PD). The filamentous component of LB from patients with PD contains biochemically altered alpha-synuclein. We have investigated the effect of the oxidized products of catecholamines on the modification of alpha-synuclein. When alpha-synuclein was incubated with the oxidized 3,4-dihydroxyphenylalanine (L-DOPA) or dopamine, the protein was induced to be aggregated. The oxidized catecholamine-mediated alpha-syhuclein aggregation was enhanced by copper ion. Radical scavengers, azide and N-acetyl cysteine significantly prevented the oxidized catecholamine-mediated alpha-synuclein aggregation. The results suggest that free radical may play a role in alpha-synuclein aggregation. Exposure of alpha-synuclein to the oxidized products of catecholamines led to the formation of dityrosine. Antioxidant dipeptides carnosine, homocarnosine and anserine significantly protected alpha-synuclein from the aggregation induced by the oxidized products of catecholamines.

Khan FH, Sen T, Maiti AK, Jana S, Chatterjee U, Chakrabarti S. 2005. Inhibition of rat brain mitochondrial electron transport chain activity by dopamine oxidation products during extended in vitro incubation: Implications for Parkinson's disease. Biochimica Et Biophysica Acta-Molecular Basis of Disease 1741(1-2):65-74.

Abstract: Several studies on mitochondrial functions following brief exposure (5-15 min) to dopamine (DA) in vitro have produced extremely variable results. In contrast, this study demonstrates that a prolonged exposure (up to 2 h) of disrupted or lysed mitochondria to DA (0.1-0.4 mM) causes a remarkable and dose-dependent inhibition of complex I and complex IV activities. The inhibition of complex I and complex IV activities is not prevented by the antioxidant enzyme catalase (0.05 mg/ml) or the metal-chelator diethylenetriaminepentaacetic acid (0.1 mM) or the hydroxyl radical scavengers like mannitol (20 mM) and dimethyl sulphoxide (20 mM) indicating the non-involvement of OH radicals and Fenton's chemistry in this process. However, reduced glutathione (5 mM), a quinone scavenger, almost completely abolishes the DA effect on mitochondrial complex I and complex IV activities, while tyrosinase (250 units/ml) which catalyses the conversion of DA to quinone products dramatically enhances the former effect. The results suggest the predominant involvement of quinone products instead of reactive oxygen radicals in long-term DA-mediated inactivation of complex I and complex TV. This is further indicated from the fact that significant amount of quinones and quinoprotein adducts (covalent adducts of reactive quinones with protein thiols) are formed during incubation of mitochondria with DA. Monoamine oxidase A (MAO-A) inhibitor clorgyline also provides variable but significant protection against DA induced inactivation of complex I and complex IV activities, presumably again through inhibition of quinoprotein formation. Mitochondrial ability to reduce tetrazolium dye 3-[4,5-dimethylthiazol-2-yl]-2,5-diphenyltetrazolium bromide (MTT) in presence of a respiratory substrate like succinate (10 mM) is also reduced by nearly 85% following 2 h incubation with 0.4 mM DA. This effect of DA on mitochondrial function is also dose-dependent and presumably mediated by quinone products of DA oxidation. The mitochondrial dysfunction induced by dopamine during extended periods of incubation as reported here have important implications in the context of dopaminergic neuronal death in Parkinson's disease (PD). (c) 2005 Elsevier B.V. All rights reserved.

Khan A, Ashcroft AE, Higenell V, Korchazhkina OV, Exley C. 2005. Metals accelerate the formation and direct the structure of amyloid fibrils of NAC. J Inorg Biochem 99(9):1920-1927.
Abstract: Non-beta amyloid component of Alzheimer's disease amyloid or NAC is a highly amyloidogenic peptide consisting of 35 amino acids which was first identified associated with senile plaques in the Alzheimer's disease brain. It is a fragment of the presynaptic protein alpha-synuclein and, as such, it is implicated in the aetiologies of both Alzheimer's (AD) and Parkinson's (PD) disease. Metals are involved in the aggregation of amyloidogenic peptides such as beta amyloid (A P), British amyloid peptide (ABri) and (X-synuclein though nothing is yet known about how they might influence the aggregation of NAC. We show herein that NAC will form beta-pleated conformers at a peptide concentration of only 2.0 mu M and that metals, and Zn(II) and Cu(II) in particular, accelerate the formation of these fibrils. Cu(II) and Zn(II) did not influence the diameter or general structure of the fibrils which were formed though many more shorter fibrils were observed in their presence and these shorter fibrils were highly thioflavin T positive and they were efficient catalysts of the redox cycling of added Fe(II). By way of contrast, beta-pleated conformers of NAC which were formed in the presence of Al(III) showed much lower levels of thioflavin T fluorescence and were poorer catalysts of the redox cycling of added Fe(II) and these properties were commensurate with an increased abundance of a novel amyloid morphology which consisted of twisted fibrils with a periodicity of about 100 nm. These spirals of twisted fibrils were especially abundant in the presence of added AI(III) and it is speculated that NAC binding of Al(III) may be important in their formation and subsequent stability. (c) 2005 Elsevier Inc. All rights reserved.

Keller J, Owens CT, Lai JCK, Devaud LL. 2005. The effects of 17 beta-estradiol and ethanol on zinc- or manganese-induced toxicity in SK-N-SH cells. Neurochem Int 46(4):293-303.

Abstract: Serious neurodegenerative disorders are increasingly prevalent in our society and excessive oxidative stress may be a key mediator of neuronal cell death in many of these conditions. A variety of metals, such as manganese and zinc, are essential trace elements but can reach localized toxic concentrations through various disease processes or environmental exposures and have been implicated as having a role in neurodegeneration. Both manganese and zinc exist as bivalent cations and are essential cofactors/activators for numerous enzymes. Evidence suggests one action of these metals, when concentrated beyond physiological levels, may be to inhibit cellular energy production, ultimately leading to increased radical formation. Our studies were undertaken to directly investigate the toxic effects of manganese and zinc in an immortalized neuronal-like cell line (SK-N-SH) by testing interactions with the antioxidant, 17beta-estradiol, and the neurotoxin, ethanol. Employing undifferentiated SK-N-SH cells, we found that these metals caused biphasic effects, enhancing cell proliferation at low doses and inducing cell death at higher doses. Zinc was both more efficacious and more potent than manganese in enhancing growth and in causing cell death. 17beta-Estradiol and ethanol enhanced the proliferative actions of zinc and manganese across a wide concentration range. Furthermore, co-treatment with either 17beta-estradiol or ethanol afforded protection against manganese-, but not zinc-induced toxicity. Finally, combined administration of 17beta-estradiol and ethanol to SK-N-SH cells resulted in both a loss of growth enhancement and protective properties that were observed when these substances were administered individually. We also noted that the toxic effects occurred more rapidly from zinc than manganese exposure. Taken together, these data suggest that oxidative stress likely has a role in cell death resulting from toxic exposure to either zinc or manganese, but there is a difference in the precise mechanism of their effects. (C) 2004 Elsevier Ltd. All rights reserved.

Kanthasamy AG, Kitazawa M, Kanthasamy A, Anantharam V. 2005. Dieldrin-induced neurotoxicity: Relevance to Parkinson's disease pathogenesis. Neurotoxicology 26(4):701-719.
Abstract: Parkinson's disease (PD) is increasingly recognized as a neurodegenerative disorder strongly associated with environmental chemical exposures. Recent epidemiological data demonstrate that environmental risk factors may play a dominant role as compared to genetic factors in the etiopathogenesis of idiopathic Parkinson's disease. Identification of key genetic defects such as alpha-synuclein and parkin mutations in PD also underscores the important role of genetic factors in the disease. Thus, understanding the interplay between genes and environment in PD may be critical to unlocking the mysteries of this 200-year-old neurodegenerative disease. Pesticides and metals are the most common classes of environmental chemicals that promote dopaminergic degeneration. The organochlorine pesticide dieldrin has been found in human PD postmortem brain tissues, suggesting that this pesticide has potential to promote nigral cell death. Though dieldrin has been banned, humans continue to be exposed to the pesticide through contaminated dairy products and meats due to the persistent accumulation of the pesticide in the environment. This review summarizes various neurotoxic studies conducted in both cell culture and animals models following dieldrin exposure and discusses their relevance to key pathological mechanisms associated with nigral dopaminergic degeneration including oxidative stress, mitochondrial dysfunction, protein aggregation, and apoptosis. (c) 2004 Elsevier Inc. All rights reserved.

Josephs KA, Ahlskog JE, Klos KJ, Kumar N, Fealey RD, Trenerry MR, Cowl CT. 2005. Neurologic manifestations in welders with pallidal MRI T1 hyperintensity. Neurology 64(12):2033-2039.
Abstract: Background: Neurologic symptoms have been attributed to manganese fumes generated during welding. Increased T1 MRI signal in the basal ganglia is a biologic marker of manganese accumulation. Recent studies have associated welding and parkinsonism, but generally without MRI corroboration. Objective: To characterize the clinical and

neuropsychological features of patients with MRI basal ganglia T1 hyperintensity, who were ultimately diagnosed with neurotoxicity from welding fumes. Methods: The medical records of welders referred to the Department of Neurology with neurologic problems and basal ganglia T1 hyperintensity were reviewed. Results: All eight patients were male career welders with increased T1 basal ganglia signal on MRI of the brain. Several different clinical syndromes were recognized: a parkinsonian syndrome (three patients), a syndrome of multifocal myoclonus and limited cognitive impairment (two patients), a mixed syndrome with vestibular-auditory dysfunction (two patients), and minor subjective cognitive impairment, anxiety, and sleep apnea (one patient). Neuropsychometric testing suggested subcortical or frontal involvement. Inadequate ventilation or lack of personal respiratory protection during welding was a common theme. Conclusions: Welding without proper protection was associated with syndromes of parkinsonism, multifocal myoclonus, mild cognitive impairment, and vestibular - auditory dysfunction. The MRI T1 hyperintensity in the basal ganglia suggests that these may have been caused by manganese neurotoxicity.

Jones S, Gibb AJ. 2005. Functional NR2B- and NR2D-containing NMDA receptor channels in rat substantia nigra dopaminergic neurones. Journal of Physiology-London 569(1):209-221.
Abstract: NMDA receptors regulate burst firing of dopaminergic neurones in the substantia nigra pars compacta (SNc) and may contribute to excitotoxic cell death in Parkinson's disease (PD). In order to investigate the subunit composition of functional NMDA receptors in identified rat SNc dopaminergic neurones, we have analysed the properties of individual NMDA receptor channels in outside-out patches. NMDA (100 nm) activated channels corresponding to four chord conductances of 18, 30, 41 and 54 pS. Direct transitions were observed between all conductance levels. Between 18 pS and 41 pS conductance levels, direct transitions were asymmetric, consistent with the presence of NR2D-containing NMDA receptors. Channel activity in response to 100 nm or 200 mu m NMDA was not affected by zinc or TPEN (N,N,N ',N '-tetrakis-[2-pyridylmethyl]-ethylenediamine), indicating that SNc dopaminergic neurones do not contain functional NR2A subunits. The effect of the NR2B antagonist ifenprodil was complex: 1 mu m ifenprodil reduced open probability, while 10 Am reduced channel open time but had no effect on open probability of channels activated by 100 nm NMDA. When the concentration of NMDA was increased to 200 mu m, ifenprodil (10 mu m) produced the expected reduction in open probability. These results indicate that NR2B subunits are present in SNc dopaminergic neurones. Taken together, these findings indicate that NR2D and NR2B subunits form functional NMDA receptor channels in SNc dopaminergic neurones, and suggest that they may form a triheteromeric NMDA receptor composed of NR1/NR2B/NR2D subunits.

Jankovic J. 2005. Searching for a relationship between manganese and welding and Parkinson's disease. Neurology 64(12):2021-2028.
Abstract: Research into the causes of Parkinson disease (PD) has accelerated recently with the discovery of novel gene mutations. The majority of PD cases, however, remain idiopathic and in those cases environmental causes should be considered. Several recent reports have focused on welding and manganese toxicity as potential risk factors for parkinsonism and some have even proposed that welding is a risk factor for PD. The controversy has stimulated this review, the primary aim of which is to critically and objectively examine the evidence or lack of evidence for a relationship among welding, manganese, parkinsonism, and PD.

Izumi Y, Sawada H, Yamamoto N, Kume T, Katsuki H, Shimohama S, Akaike A. 2005. Iron accelerates the conversion of dopamine-oxidized intermediates into melanin and provides protection in SH-SY5Y cells. J Neurosci Res 82 (1):126-137.
Abstract: Parkinson's disease (PD) is characterized by the selective loss of dopaminergic neurons in the substantia nigra (SN), and it has been

suggested that dopamine is one of the main endogenous toxins in the genesis of PD. We demonstrated that thiol antioxidants (the reduced form of glutathione, N-acetyl-L-cysteine, and L-cysteine), which conjugate with one dopamine oxidation intermediate, o-quinone, provided almost complete protection from dopamine-mediated toxicity in SH-SY5Y, a human neuroblastoma cell line. In contrast, catalase partially provided protection against cell death caused by dopamine. These data suggest that the generation of dopamine oxidation intermediates, rather than hydrogen peroxide, plays a pivotal role in dopamine-induced toxicity. Iron accumulated in the SN of patients with PD can cause dopaminergic neuronal degeneration by enhancing oxidative stress. However, we found that iron reduced the total amounts of dopamine oxidation intermediates and enhanced the formation of melanin, a final product of dopamine oxidation. Also, addition of iron inhibited dopamine-induced cytotoxicity. These results suggest that iron can provide protection when it accelerates the conversion of dopamine oxidation intermediates. (C) 2005 Wiley-Liss, Inc.

Izumi Y, Sawada H, Sakka N, Yamamoto N, Kume T, Katsuki H, Shimohama S, Akaike A. 2005 . p-quinone mediates 6-hydroxydopamine-induced dopaminergic neuronal death and ferrous iron accelerates the conversion of p-quinone into melanin extracellularly. J Neurosci Res 79(6):849-860.
Abstract: Parkinson's disease (PD) is characterized by the selective loss of dopaminergic neurons in the substantia nigra (SN). 6-Hydroxydopamine (6-OHDA), a dopaminergic neurotoxin, is detected in human brains and the urine of PD patients. Using SH-SY5Y, a human neuroblastoma cell line, we demonstrated that 6-OHDA toxicity was determined by the amount of p-quinone produced in 6-OHDA auto-oxidation rather than by reactive oxygen species (ROS). Glutathione (GSH), which conjugated with p-quinone, provided significant protection whereas catalase, which detoxified hydrogen peroxide and superoxide anions, failed to block cell death caused by 6-OHDA. Although iron accumulated in the SN of patients with PD can cause dopaminergic neuronal degeneration by enhancing oxidative stress, we found that extracellular ferrous iron promoted the formation of melanin and reduced the amount of p-quinone. The addition of ferrous iron to the culture medium inhibited caspase-3 activation and apoptotic nuclear morphologic changes and blocked 6-OHDA-induced cytotoxicity in SH-SY5Y cells and primary cultured mesencephalic dopaminergic neurons. These data suggested that generation of p-quinone played a pivotal role in 6-OHDA-induced toxicity and extracellular iron in contrast to intracellular iron was protective rather than harmful because it accelerated the conversion of p-quinone into melanin. (C) 2005 Wiley-Liss, Inc.

Ide-Ektessabi A, Rabionet M. 2005. The role of trace metallic elements in neurodegenerative disorders: Quantitative analysis using XRF and XANES spectroscopy. Analytical Sciences 21(7):885-892.
Abstract: The present paper focuses on the analysis of trace metallic elements and their role in neurodegenerative disorders. The use of synchrotron radiation microbeams allows investigation of pathological tissues from Alzheimer's disease, Parkinson's disease and Amyotrophic lateral sclerosis cases in a nondestructive manner and at cellular level. By employing X-ray absorption near edge structure (XANES) technique, the chemical state of the investigated elements can be determined, while energy-selective X-ray fluorescence spectroscopy provides the spatial distribution of each element in each oxidative state selectively. The investigated tissues (derived from human, monkey and mouse specimens) show distinct imbalances of metallic elements such as Zn and Cu as well as Fe2+/Fe3+ redox pair, which point to oxidative stress as a crucial factor in the development or progress of these neurodegenerative diseases.

Huang E, Ong WY. 2005. Distribution of ferritin in the rat hippocampus after kainate-induced neuronal injury. Exp Brain Res 161 (4):502-511.
Abstract: A gradual increase in iron occurs in the lesioned hippocampus after neuronal injury induced by the excitotoxin kainate, and the present study was carried out to investigate whether this increase in iron might be

associated with changes in expression of the iron binding protein, ferritin. An increase in ferritin immunoreactivity was observed in glial cells of the hippocampus, as early as three days after intracerebroventricular injections of kainate. The number of ferritin positive cells peaked four weeks after the kainate injection, and decreased eight and twelve weeks after injection. They were found to be mostly microglia and oligodendrocytes by double immunofluorescence labeling with glial markers. A number of ferritin-labeled endothelial cells were also observed via electron microscopy. The decline in ferritin immunoreactivity four weeks after the injection of kainate is accompanied by an increase in the number of ferric and ferrous iron positive cells in the lesioned tissue. A substantial non-overlap between ferritin and iron-containing cells was observed. In particular, spherical ferric or ferrous iron-laden cells in the degenerating hippocampus were unlabeled for ferritin for long time periods after the kainate injection. An increase in iron, together with a reduced expression of iron binding proteins such as ferritin at long time intervals after kainate lesions, could result in a relative decrease in ferritin-induced ferroxidase activity and the presence of some of the iron in the ferrous form. It is postulated that this may contribute to chronic neuronal injury, following acute kainate-induced neurodegeneration.

Hochstrasser H, Tomiuk J, Walter U, Behnke S, Spiegel J, Kruger R, Georg B, Riess O, Berg D. 2005. Functional relevance of ceruloplasmin mutations in Parkinson's Disease. FASEB J 19(11):1851-1853.
Abstract: Increased iron levels of the substantia nigra and the discovery of ceruloplasmin mutations in patients with Parkinson's disease (PD) imply impaired iron metabolism in this neurodegenerative disorder. Ceruloplasmin has ferroxidase activity oxidizing iron(II) to iron(III). In the present study, we analyzed the amount of ceruloplasmin, iron, ferritin, and transferrin and the ceruloplasmin ferroxidase activity in serum of patients with the diagnosis of PD carrying the ceruloplasmin mutations I63T, D544E, and R793H. The impact of these missense mutations on the biosynthesis of holo-ceruloplasmin was investigated in cell culture experiments. Functional relevance was found for the ceruloplasmin mutations I63T and D544E. In vivo, the I63T mutation resulted in half the normal ceruloplasmin concentration and markedly reduced ferroxidase activity in serum from a heteroallelic PD patient. In cell culture, the I63T glycosylphosphatidylinositol (GPI)-linked ceruloplasmin isoform was retained in the endoplasmatic reticulum of human embryonic kidney cells. Furthermore, the D544E polymorphism resulted in significantly reduced serum ceruloplasmin levels and ferroxidase activity in heteroallelic patients and in expression of mainly apo-ceruloplasmin in cell culture. Our studies indicate that altered activity of ceruloplasmin may present a vulnerability factor for iron induced oxidative stress in PD.

Hirata Y, Nagatsu T. 2005. Rotenone and CCCP inhibit tyrosine hydroxylation in rat striatal tissue slices. Toxicology 216(1):9-14.
Abstract: Complex I inhibition has been implicated in the neurotoxicity of MPTP and rotenone, which reproduce a neurochernical and neuropathological feature of Parkinson's disease in experimental animals. Previous studies performed in rat striatal slices have shown that dopaminergic neurotoxins, MPTP and manganese, inhibit tyrosine hydroxylation, a rate-limiting step of dopamine biosynthesis. In this study, we examined the effect of mitochondrial toxins such as rotenone and carbonyl cyanide 3-chlorophenylhydrazone (CCCP) on tyrosine hydroxylation in rat striatal slices. Rotenone and CCCP inhibited DOPA formation with an accompanying decrease in ATP and increase in lactate of rat striatal slices during 1 h incubation. Furthermore, rotenone reduced dopamine (DA), dihydroxyphenyl acetic acid (DOPAC) and homovanillic acid (HVA) levels in PC 12 cells after 20 h incubation. These results suggest that tyrosine hydroxylation is inhibited in dopaminergic neurons soon after exposure to sub-micromolar concentrations of rotenone and CCCP, leading to dopamine depletion. (c) 2005 Elsevier Ireland Ltd. All rights reserved.

Hikita T, Abe K, Sakoda S, Tanaka H, Murase K, Fujita N. 2005. Determination of

transverse relaxation rate for estimating iron deposits in central nervous system. Neurosci Res 51(1):67-71.
Abstract: To determine the amount of iron deposits in the basal ganglia, we examined 13 healthy volunteers with a 1.5 T MRI system using three transverse relaxation rates measured with two sequences. The transverse relaxation rates comprise the reversible contribution (R2') and irreversible contribution (R2) to a phase-reversal 180degrees-pulse sequence. The transverse relaxation rates with the estimated iron indicated that both R2 and R2' had a robust relationship with brain iron level as determined from published post mortem data. This was the case only when the analyses are limited to the subcortical gray matter regions, however. R2' was affected by macroscopic magnetic field inhomogeneity arising from the skull bases, so that it was less robust for estimating the amount of iron deposits in the basal ganglia. (C) 2004 Elsevier Ireland Ltd and the Japan Neuroscience Society. All rights reserved.

Hermosura MC, Nayakanti H, Dorovkov MV, Calderon FR, Ryazanov AG, Haymer DS, Garruto RM. 2005. A TRPM7 variant shows altered sensitivity to magnesium that may contribute to the pathogenesis of two Guamanian neurodegenerative disorders. Proc Natl Acad Sci U S A 102(32): 11510-11515.
Abstract: Guamanian amyotrophic lateral sclerosis (ALS-G) and parkinsonism dementia (PD-G) have been epidemiologically linked to an environment severely deficient in calcium (Ca2+) and magnesium (Mg2+). Transient receptor potential melastatin 7 (TRPM7) is a bifunctional protein containing both channel and kinase domains that has been proposed to be involved in the homeostatic regulation of intracellular Ca2+, Mg2+, and trace metal ion concentration. There is evidence that TRPM7 is constitutively active and that the number of available channels is dependent on intracellular free Mg2+ levels. We found a TRPM7 variant in a subset of ALS-G and PD-G patients that produces a protein with a missense mutation, T14821. Recombinant T14821 TRPM7 exhibits the same kinase catalytic activity as WT TRPM7. However, heterologously expressed T14821 TRPM7 produces functional channels that show an increased sensitivity to inhibition by intracellular Mg2+. Because the incidence of ALS-G and PD-G has been associated with prolonged exposure to an environment severely deficient in Ca2+ and Mg2+, we propose that this variant TRPM7 allele confers a susceptibility genotype in such an environment. This study represents an initial attempt to address the important issue of gene-environment interactions in the etiology of these diseases.

Hardy PA, Gash D, Yokel R, Andersen A, Ai Y, Zhang ZM. 2005. Correlation of R2 with total iron concentration in the brains of rhesus monkeys. J Magn Reson Imaging 21(2):118-127.
Abstract: Purpose: To estimate the relationship between R-2 = 1/T-2 as measured with a double echo spin echo sequence and total iron concentration in gray matter structures in the brains of aging rhesus monkeys. Materials and Methods: Using a 1.5-T magnetic resonance (MR) imager, we collected double echo spin echo images of the brains of 12 female rhesus monkeys aged between 9 and 23 Years. From the double echo images. the transverse relaxation rate R-2 = 1/T-2 was calculated in selected gray matter regions. After the animals were euthanized, their brains Were excised and tissue punches were taken of the substantia nigra, globus pallidus. and gray matter regions of the cerebellum. Some of the tissue punches were assayed for total iron using atomic absorption spectroscopy. Results: The range of, tissue iron concentration spanned from 15 to 450 mug/g wet weight with the highest levels in the globus pallidus and the lowest levels in the cerebellum. The results show that R-2 was highly correlated with the total iron concentration and that the relationship between R-2 and tissue iron concentration appeared to depend upon the iron concentration. For concentrations above approximately 150 mug/g wet weight, R-2 increased with a sensitivity of 0.0484 +/- 0.0023 second(-1) (mug/g)(-1). In contrast. where the iron concentration was below 150 mug/g, R-2 increased at 0.0013 +/- 0.0073 second(-1)(mug/g)

(-1). The bilinear behavior may reflect changes with age in the relative amounts of iron distributed diffusely and in granular form in the globus pallidus and substantia nigra. Histological sections of the tissues stained for iron and ferritin support this hypothesis and indicate that the distribution of ferritin is similar to the distribution of iron. Conclusion: This study reaffirms the value of measuring the MR relaxation rate R-2 for a noninvasive estimate of iron content in the brain and identified limitations in the relationship at low tissue iron concentrations.

Halliday GM, Ophof A, Broe M, Jensen PH, Kettle E, Fedorow H, Cartwright MI, Griffiths FM, Shepherd CE, Double KL. 2005. alpha-Synuclein redistributes to neuromelanin lipid in the substantia nigra early in Parkinson's disease. Brain 128:2654-2664.
Abstract: The distribution and tempo of neuronal loss in Parkinson's disease correlates poorly with the characteristic and more widely spread intracellular changes associated with the disease process (Lewy bodies and Lewy neurites). To determine early intracellular changes in regions where cell loss is most marked (dopaminergic A9 substantia nigra) versus regions with Lewy bodies but where cell loss is limited, we assessed 13 patients with definite Parkinson's disease at various disease stages in comparison with controls. Using immunohistochemistry for alpha-synuclein, we confirmed the concentration of this protein in the soma of normal A9 neurons and in Lewy body pathology in brainstem catecholamine neurons in Parkinson's disease. Analysis of the degree of cell loss in brainstem catecholamine cell groups revealed that only the A9 substantia nigra had consistent significant cell loss early in the disease course with greater A9 cell loss correlating with increasing disease duration. To assess the earliest intracellular changes differentiating neurons more likely to degenerate, pigmented A9 and A10 neurons with and without obvious pathology were targeted, cell size and pigment density measured, and intracellular changes in alpha-synuclein location and lipid components analysed at both the light and electron microscope levels. There were no changes observed in healthy A10 neurons in Parkinson's disease compared with controls. Pigmented A9 neurons in later stages of degeneration with obvious Lewy body formation had a significant reduction in intracellular pigment, as previously described. In contrast, A9 neurons of normal morphological appearance and no characteristic pathology in Parkinson's disease exhibited significantly increased pigment density associated with a concentration of alpha-synuclein to the lipid component of the pigment and a loss of associated cholesterol. These changes in vulnerable but apparently healthy A9 neurons occurred without any change in cell size or in the amount of intracellular pigment compared with controls. The increase in pigment density is consistent with previously reported increases associated with oxidation and iron loading, reactions known to precipitate alpha-synuclein. The selectivity of the changes observed in A9 nigral neurons suggests that these early intracellular changes predispose these neurons to more rapid cell loss in Parkinson's disease. The increased concentration of neuronal alpha-synuclein and pigment in normal A9 neurons may already predispose these neurons to precipitate alpha-synuclein around pigment-associated lipid under oxidative conditions. Overall, these changes may trigger a cascade of events leading to larger intracellular aggregates of alpha-synuclein and the dispersement of protective pigment to precipitate cell death in Parkinson's disease.

Haba-Rubio J, Staner L, Petiau C, Erb G, Schunck T, Macher JP. 2005. Restless legs syndrome and low brain iron levels in patients with haemochromatosis. J Neurol Neurosurg Psychiatry 76(7):1009-1010.
Abstract: Regional brain iron levels of two patients with haemochromatosis and severe restless legs syndrome (RLS) were assessed using R2' magnetic resonance imaging (MRI) sequences in both patients and in nine healthy controls. R2' relaxation rates in the patients were decreased in the substantia nigra, red nucleus, and pallidum when compared with the controls. These results indicate that local brain iron deficiency may occur in patients with haemochromatosis and suggest a role for brain iron

metabolism in the pathophysiology of RLS.

Haacke EM, Chengb NYC, House MJ, Liu Q, Neelavalli J, Ogg RJ, Khan A, Ayaz M, Kirsch W, Obenaus A. 2005. Imaging iron stores in the brain using magnetic resonance imaging. Magn Reson Imaging 23(1):1-25.
Abstract: For the last century, there has been great physiological interest in brain iron and its role in brain function and disease. It is well known that iron accumulates in the brain for people with Huntington's disease, Parkinson's disease, Alzheimer's disease, multiple sclerosis, chronic hemorrhage, cerebral infarction, anemia, thalassemia, hemochromatosis, Hallervorden-Spatz, Down syndrome, AIDS and in the eye for people with macular degeneration. Measuring the amount of nonheme iron in the body may well lead to not only a better understanding of the disease progression but an ability to predict outcome. As there are many forms of iron in the brain, separating them and quantifying each type have been a major challenge. In this review, we present our understanding of attempts to measure brain iron and the potential of doing so with magnetic resonance imaging. Specifically, we examine the response of the magnetic resonance visible iron in tissue that produces signal changes in both magnitude and phase images. These images seem to correlate with brain iron content, perhaps ferritin specifically, but still have not been successfully exploited to accurately and precisely quantify brain iron. For future quantitative studies of iron content we propose four methods: correlating RT and phase to iron content; applying a special filter to the phase to obtain a susceptibility map; using complex analysis to extract the product of susceptibility and volume content of the susceptibility source; and using early and late echo information to separately predict susceptibility and volume content. (C) 2005 Elsevier Inc. All rights reserved.

Gunter KK, Aschner M, Miller LM, Eliseev R, Salter J, Anderson K, Hammond S, Gunter TE. 2005. Determining the oxidation states of manganese in PC12 and nerve growth factor-induced PC12 cells. Free Radic Biol Med 39(2): 164-181.
Abstract: Excessive brain Mn can produce toxicity with symptoms resembling parkinsonism. This syndrome, called "manganism," correlates with loss of dopamine in the striatum and cell death in the striatum and globus pallidus. A common hypothesis is that cell damage in Mn toxicity is caused by oxidation of important cell components by Mn3+. Determination of the amount of Mn3+ present, under a range of conditions, in neuronal cells and brain mitochondria represents an important step in evaluating the "damage through oxidation by Mn3+ hypothesis." In an earlier paper we used X-ray absorption near-edge structure (XANES) spectroscopy to determine the amount of Mn2+ and Mn3+ in brain mitochondria under a range of conditions. Here we extend the study to investigate the evidence for formation of Mn3+ through oxidation of Mn2+ by ROS in PC12 cells and in PC12 cells induced with nerve growth factor (NGF) to display a phenotype more like that of neurons. Although the results suggest that very small amounts of Mn3+ might be present at low Mn levels, probably in Mn superoxide dismutase, Mn3+ is not stabilized by complex formation in these cells and therefore does not accumulate to detectable amounts. (C) 2005 Elsevier Inc. All rights reserved.

Gun J, Ekeltchik I, Lev O, Shelkov R, Melman A. 2005. Bis-(hydroxyamino) triazines: highly stable hydroxylamine-based ligands for iron(III) cations. Chemical Communications (42):5319-5321.
Abstract: Bis-(hydroxyamino)triazines (BHTs) constitute a new, general and highly versatile group of tridentate iron(III) chelating agents exhibiting higher affinity to iron(III) than other tridentate iron(III) chelators and superior iron(III) over iron(II) selectivity compared to desferrioxamine-B (DFO), EDTA as well as other tridentate ligands.

Gossuin Y, Muller RN, Gillis P, Bartel L. 2005. Relaxivities of human liver and spleen ferritin. Magn Reson Imaging 23(10):1001-1004.
Abstract: Ferritin, the iron-storing protein of mammals, is known to darken

T-2-weighted magnetic resonance images. This darkening can be used to noninvasively measure an organ's iron content. Significant discrepancies exist between T-2 data obtained with ferritin-containing tissues and with aqueous solutions of horse spleen ferritin (HSF). The NMR properties of stable human ferritin have never been studied in aqueous solutions. Relaxometry results on human liver and spleen ferritin are reported here, showing that the relaxation induced in aqueous solutions by human ferritins is comparable to that induced by HSF. As a consequence, the differences between ferritin-containing human tissues and ferritin solutions cannot be attributed to different NMR properties of human and horse ferritins, but probably to a clustering of the protein in vivo. (c) 2005 Elsevier Inc. All rights reserved.

Gossuin Y, Hautot D, Muller RN, Pankhurst Q, Dobson J, Morris C, Gillis P, Collingwood J. 2005. Looking for biogenic magnetite in brain ferritin using NMR relaxometry. NMR Biomed 18(7):469-472.
Abstract: Mammalian cellular iron is stored inside the multisubunit protein ferritin. normally taking the structure of a ferrihydrite-like mineral core. It has been suggested that biogenic magnetite, which has been detected in the brain and may be related to neurodegenerative diseases such as Alzheimer's and Parkinson's diseases, could initially form in ferritin. Indeed, as ferritin is present in the brain, the ferrihydrite core could be a precursor for biogenic magnetite formation 19 particularly in cases where the normal functioning of the ferritin protein is disrupted. In this work, NMR relaxometry was used to detect magnetite inside samples of ferritin extracted from normal and Alzheimer-diseased brains. The method was first calibrated with different fractions of horse spleen ferritin and synthetic magnetite particles. The relaxometry results suggest that the proportion of iron contained in brain ferritin in the form of well-crystallized magnetite instead of ferrihydrite must be < 1%, which is much less than that reported for 'magnetite-like' phase in recent transmission electron microscopy studies of similar samples. Consequently, the magnetization of this 'magnetite-like' phase must be very low compared with that of magnetite. Copyright (c) 2005 John Wiley & Sons, Ltd.

Goldschmith A, Infante C, Leiva J, Motles E, Palestini M. 2005. Interference of chronically ingested copper in long-term potentiation (LTP) of rat hippocampus. Brain Res 1056(2):176-182.
Abstract: The objective of our study was to find the evidence of copper interaction in LTP, motivated by copper involvement in neurodegenerative illness, like Parkinson, Alzheimer and Amyotrophic Lateral Sclerosis, and we initiated the study of this element in the LTP. For this purpose we used hippocampus slices of rats chronically consuming copper dissolved in water (CuDR: n = 26) and non-copper-consuming rats (CR; n = 20). The CuDR rats received 8-10 mg/day during 20-25 days. Electrophysiological tests showed absence of LTP in CuDR slices, contrary to CR slices. The stimulus-response test applied before and after LTP showed significant increases of synaptic potential in the CR group. This did not occur in the CuDR group, except for the initial values, which probably seem associated to an early action of copper, The paired-pulse (PP) test, applied to CR and CuDR prior to tetanic stimulation, showed a significant reduction in PP, for the 20-, 30- and 50-ms intervals in CuDR. At the end of the experiments, copper concentration was 54.2 times higher in CuDR slices, compared to the concentration present in CR slices. Our results show that copper reduces synaptic sensibility and also the facilitation capability. These effects represent a significant disturbance in the plasticity phenomenon associated with learning and memory. (c) 2005 Elsevier B.V. All rights reserved.

Glaser CB, Yamin G, Uversky VN, Fink AL. 2005. Methionine oxidation, alpha-synuclein and Parkinson's disease. Biochimica Et Biophysica Acta-Proteins and Proteomics 1703(2):157-169.
Abstract: The aggregation of normally soluble alpha-synuclein in the dopaminergic neurons of the substantia nigra is a crucial step in the pathogenesis of Parkinson's disease. Oxidative stress is believed to be a contributing factor in this disorder. Because it lacks Trp and Cys residues,

mild oxidation of alpha-synuclein in vitro with hydrogen peroxide selectively converts all four methionine residues to the corresponding sulfoxides. Both oxidized and non-oxidized alpha-synucleins have similar unfolded conformations; however, the fibrillation of alpha-synuclein at physiological pH is completely inhibited by methionine oxidation. The inhibition results from stabilization of soluble oligomers of Met-oxidized alpha-synuclein. Furthermore, the Met-oxidized protein also inhibits fibrillation of unmodified alpha-synuclein. The degree of inhibition of fibrillation by Met-oxidized alpha-synuclein is proportional to the number of oxidized methionines. However, the presence of metals can completely overcome the inhibition of fibrillation of the Met-oxidized alpha-synuclein. Since oligomers of aggregated alpha-synuclein may be cytotoxic, these findings indicate that both oxidative stress and environmental metal pollution could play an important role in the aggregation of alpha-synuclein, and hence possibly Parkinson's disease. In addition, if the level of Met-oxidized alpha-synuclein was under the control of methionine sulfoxide reductase (Msr), then this could also be factor in the disease. (C) 2004 Elsevier B.V. All rights reserved.

Giorelli M, Livrea P, Trojano M. 2005. Dopamine fails to regulate activation of peripheral blood lymphocytes from multiple sclerosis patients: Effects of IFN-beta. J Interferon Cytokine Res 25(7):395-406.
Abstract: The neurotransmitter dopamine counteracts T cell functions through its specific receptor subtype D5R but favors T cell proliferation and adhesion when acting on D3R. We found diminished mRNA and protein levels of D5R, but not of D3R, in peripheral blood mononuclear cells (PBMCs) from untreated multiple sclerosis (MS) patients. Dopamine reduced T cell proliferation, secretion of interferon-gamma (IFN-gamma), and production of matrix metalloproteinase-9 (MMP-9) mRNA in PBMCs from controls but not from MS patients. By contrast, reduced levels of D3R and renewed dopamine-associated regulatory functions were found in PBMCs from IFN-beta treated MS patients. Failure of the dopaminergic system of lymphocytes may lessen the threshold of T cell activation and sustain the pathogenic cascade of MS.

Gimsa J, Habel B, Schreiber U, Van Rienen U, Strauss U, Gimsa U. 2005. Choosing electrodes for deep brain stimulation experiments - electrochemical considerations. J Neurosci Methods 142(2):251-265.
Abstract: Deep brain stimulation (DBS) is a therapy of movement disorders including Parkinson's disease (PD). Commercially available electrodes for animal models of Parkinson's disease vary in geometry and material. We characterized such electrodes and found a drift in their properties within minutes and up to about 60 h after immersion in cell culture medium, both with and without a stimulation signal. Electrode properties could largely be restored by proteolytic treatment for platinum/iridium electrodes but not for stainless steel ones. Short-term drift and irreversible aging could be followed by impedance measurements. Aging was accompanied by metal corrosion and erosion of the plastic insulation. For both materials, the degradation rates depended on the current density at the electrode surfaces. Fourier analysis of the DBS pulse (60 mus, repetition rate 130Hz) revealed harmonic frequencies spanning a band of more than three decades, with significant harmonics up to the MHz range. The band is located in a window imposed by electrode processes and capacitive cell membrane bridging at the low and high frequency ends, respectively. Even though electrode processes are reduced at higher frequencies they only vanish above 1 MHz and cannot be avoided. Therefore, the use of inert electrode materials is of special importance. The neurotoxicity of iron makes avoiding stainless steel electrodes imperative. Future developments need to avoid the use of corrosive materials and current density hot spots at the electrode surface, and to reduce low frequency components in the DBS pulses in order to diminish electrode processes. (C) 2004 Elsevier B.V. All rights reserved.

Gille G, Reichmann H. 2005. Update on etiopathogenesis of idiopathic Parkinson's syndrome. Aktuelle Neurologie 32: S75-S87.

Abstract: Intensive research activities are carried out to elucidate the causes underlying idiopathic Parkinson's disease. In recent years, significant progress has been achieved concerning the pathogenetic mechanisms and the hereditary cases have contributed valuable insights. This short review outlines the putative toxic mechanisms connected with mutations of the PARK genes, and summarizes other potentially important factors like environmental causes, inflammation, proteasome, mitochondrial dysfunction, iron toxicity and neuromelanin, dopamine, glutamate and excitotoxicity. The controversial discussion about the mechanism of dopaminergic cell death (apoptosis vs. necrosis) will be followed, and finally, oxidative stress will be presented as an intersecting common pathway of all pathogenetic factors.

Gartner CE, Battistutta D, Dunne MP, Silburn PA, Mellick GD. 2005. Test-retest repeatability of self-reported environmental exposures in Parkinson's disease cases and healthy controls. Parkinsonism & Related Disorders 11 (5):287-295.
Abstract: There is substantial disagreement among published epidemiological studies regarding environmental risk factors for Parkinson's disease (PD). Differences in the quality of measurement of environmental exposures may contribute to this variation. The current study examined the test-retest repeatability of self-report data on risk factors for PD obtained from a series of 32 PD, cases recruited from neurology clinics and 29 healthy sex-, age- and residential suburb-matched controls. Exposure data were collected in face-to-face interviews using a structured questionnaire derived from previous epidemiological studies. High repeatability was demonstrated for 'lifestyle' exposures, such as smoking and coffee/tea consumption (kappas 0.70-1.00). Environmental exposures that involved some action by the person, such as pesticide application and use of solvents and metals, also showed high repeatability (kappas > 0.78). Lower repeatability was seen for rural residency and bore water consumption (kappa 0.39-0.74). In general, we found that case and control participants provided similar rates of incongruent and missing responses for categorical and continuous occupational, domestic, lifestyle and medical exposures. (C) 2005 Elsevier Ltd. All rights reserved.

Gal S, Zheng H, Fridkin M, Youdim MBH. 2005. Novel multifunctional neuroprotective iron chelator-monoamine oxidase inhibitor drugs for neurodegenerative diseases. In vivo selective brain monoamine oxidase inhibition and prevention of MPTP-induced striatal dopamine depletion. J Neurochem 95(1):79-88.
Abstract: Several multifunctional iron chelators have been synthesized from hydroxyquinoline pharmacophore of the iron chelator, VK-28, possessing the monoamine oxidase (MAO) and neuroprotective N-propargylamine moiety. They have iron chelating potency similar to desferal. M30 is a potent irreversible rat brain mitochondrial MAO-A and -B inhibitor in vitro (IC50, MAO-A, 0.037 +/- 0.02; MAO-B, 0.057 +/- 0.01). Acute (1-5 mg/kg) and chronic [5-10 mg/kg intraperitoneally (i.p.) or orally (p.o.) once daily for 14 days]in vivo studies have shown M30 to be a potent brain selective (striatum, hippocampus and cerebellum) MAO-A and -B inhibitor. It has little effects on the enzyme activities of the liver and small intestine. Its N-desmethylated derivative, M30A is significantly less active. Acute and chronic treatment with M30 results in increased levels of dopamine (DA), serotonin(5-HT), noradrenaline (NA) and decreases in DOPAC (dihydroxyphenylacetic acid), HVA (homovanillic acid) and 5-HIAA (5-hydroxyindole acetic acid) as determined in striatum and hypothalamus. In the mouse MPTP (N-methy-4-phenyl-1,2,3,6-tetrahydropyridine) model of Parkinson's disease (PD) it attenuates the DA depleting action of the neurotoxin and increases striatal levels of DA, 5-HT and NA, while decreasing their metabolites. As DA is equally well metabolized by MAO-A and -B, it is expected that M30 would have a greater DA neurotransmission potentiation in PD than selective MAO-B inhibitors, for which it is being developed, as MAO-B inhibitors do not alter brain dopamine.

Gaeta A, Hider RC. 2005. The crucial role of metal ions in neurodegeneration: the

basis for a promising therapeutic strategy. Br J Pharmacol 146(8): 1041-1059.
Abstract: The variety of factors and events involved in neurodegeneration renders the subject a major challenge. Neurodegenerative disorders include a number of different pathological conditions, which share similar critical metabolic processes, such as protein aggregation and oxidative stress, both of which are associated with the involvement of metal ions. In this review, Alzheimer's disease, Parkinson's disease and prion disease are discussed, with the aim of identifying common trends underlying these devastating neurological conditions. Chelation therapy could be a valuable therapeutic approach, since metals are considered to be a pharmacological target for the rationale design of new therapeutic agents directed towards the treatment of neurodegeneration.

Fryzek JP, Hansen J, Cohen S, Bonde JP, Llambias MT, Kolstad HA, Skytthe A, Lipworth L, Blot W, Olsen JH. 2005. A cohort study of Parkinson's disease and other neurodegenerative disorders in Danish welders. J Occup Environ Med 47(5):466-472.
Abstract: Objective. We sought to evaluate rates of hospitalizations for neurode-generative disorders in a cohort of Danish metal manufacturing employees. Methods: A retrospective cohort study was conducted from 1977 to 2002 among 27,839 mate Danish metal-manufacturing employees, with 9,817 of those employed in departments engaged in mild or stainless-steel welding and 6,163 welders. Results: The standardized hospitalization ratio and 95 % confidence intervals (Q) for Parkinson's disease were 0.9 (CI = 0.7-1.2) for men in steel-manufacturing companies, 1.0 (CI = 0.7-1.5) for men in welding departments, and 0.9 (CI = 0.4 -1.5) for welders. Observed numbers for other neurological conditions were small and not above population expectations. Analyses for time period worked, age, and duration of welding were unremarkable. Conclusions: This relatively large cohort study with long-term follow-up provides no support for the hypothesis that rates of hospitalization for Parkinson's disease or other neurological conditions are elevated under the exposure circumstances of these Danish workers.

Frigerio R, Elbaz A, Sanft KR, Peterson BJ, Bower JH, Ahlskog JE, Grossardt BR, De Andrade M, Maraganore DM, Rocca WA. 2005. Education and occupations preceding Parkinson disease - A population-based case-control study. Neurology 65(10):1575-1583.
Abstract: Objective: To investigate the association of Parkinson disease (PD) with education and occupations using a case- control study design. Methods: The authors used the medical records- linkage system of the Rochester Epidemiology Project to identify all subjects who developed PD in Olmsted County, MN, from 1976 through 1995. Each incident case was matched by age (+/- 1 year) and sex to a general population control. The authors collected information about education and occupations using two independent sources of data: a review of the complete medical records in the system and a telephone interview. Occupations were coded using the 1980 Standard Occupational Classification. Results: Subjects with 9 or more years of education were at increased risk of PD (OR = 2.0; 95% CI = 1.1 to 3.6; p = 0.02), and there was a trend of increasing risk with increasing education (test for linear trend, p = 0.02; medical records data). Physicians were at significantly increased risk of PD using both sources of occupational data. By contrast, four occupational groups showed a significantly decreased risk of PD using one source of data: construction and extractive workers (e. g., miners, oil well drillers), production workers (e. g., machine operators, fabricators), metal workers, and engineers. These associations with increased or decreased risk did not change noticeably after adjustment for education. Conclusion: Subjects with higher education and physicians have an increased risk of Parkinson disease (PD), while subjects with some occupations presumed to involve high physical activity have a decreased risk of PD.

Fries W, Netz J, Botzel K, Steinhoff B. 2005. Guideline for the assessment of driving ability of patients with neurologkal diseases. Aktuelle Neurologie 32

(6):342-350.
Abstract: Driving ability has to be assessed for the need of the patient, or because of the duty of information by the physician, or for an official statement for the drivers licence office. In the examination, motor functions of arms and legs, visual capacities, functions of the equilibrium sense have to be judged as well as mental abilities such as visual perception, attention and endurance. To check the latter validated objective psychological tests have to be used. Some of the deficits in driving ability can be compensated for by technical alterations of the vehicle in case of physical disabilities, or by careful, anticipating and danger-conscious driving in case of mental disabilities. However, homonymous hemianopia, persisting double vision, suddenly appearing vertigo attacks or visual hemineglect syndrome can not be compensated for. Each case has to be judged individually whether a concrete risk of accident emerges from the neurological disease or its symptoms. Such risk is to assume, if the physical or metal disability may cause the danger of a road accident, or if unforeseeable disturbances of consciousness may occur, or if the driver has not sufficient insight in the risks and dangers of driving. The conditions for specific diseases are discussed. In the practical examination it is recommended to do - besides standard clinical examination - an EEG, Doppler sonography of the brain supplying blood vessels and a qualified neuropsychological testing.

Forte G, Alimonti A, Violante N, Di Gregorio M, Senofonte O, Petrucci F, Sancesario G, Bocca B. 2005. Calcium, copper, iron. magnesium, silicon and zinc content of hair in Parkinson's disease. J Trace Elem Med Biol 19 (2-3):195-201.
Abstract: The aetiology of Parkinson's disease (PD) is still unknown, but some hypotheses have focused on the imbalances in body levels of metals as co-factors of risk. To assess whether hair could be a reliable marker of possible changes, calcium (Ca), copper (Cu), iron (Fe), magnesium (Mg), silicon (Si) and zinc (Zn) were determined in hair from 81 patients affected by PD and 17 age-matched controls. Care was taken to eliminate external contamination of the hair by thorough washing. Digestion of the matrix was achieved by an acid-assisted microwave procedure. Quantification of the elements was performed by inductively coupled plasma atomic emission spectrometry. Results indicated significantly lower levels of Fe in the hair of patients ($p = 0.018$) compared with controls. Ca and Mg levels were slightly lower while Zn levels were higher in patients, although these differences were not significant; neither were variations in Cu and Si. Ca and Mg were at least 1.5 times higher in females than in males in both controls and patients. In addition, Ca correlated positively with Mg in both groups and in both sexes (p-value always less than 0.03), and negatively with age in patients ($p < 0.01$). Finally, element levels did not correlate with either the duration or the severity of the disease or with anti-Parkinson treatment. (c) 2005 Elsevier GmbH. All rights reserved.

Forte G, Alimonti A, Pino A, Stanzione P, Brescianini S, Brusa L, Sancesario G, Violante N, Bocca B. 2005. Metals and oxidative stress in patients with Parkinson's disease. Ann Ist Super Sanita 41(2):189-195.
Abstract: Twenty-six metals and the oxidative status in 71 patients affected by Parkinson's disease and 44 healthy individuals were compared in order to identify potential biomarkers of the disease. In the patients, the following significant imbalances were found ($p </= 0.05$): i) in serum, an increment of Ca, Mg, Ni, Si and V, and a decrement of Cd, Co, Fe, Li, Sn, Zn and Zr; ii) in blood, raised levels of Co, Li, Ni and Si and decreased of Al, Be, Ca, Cd, Fe, Mg, Mo, Sn, Zn and Zr; iii) increased formation of oxidant species and lowered anti-oxidant capacity ($p </= 0.001$ for both). Barium, Bi, Cr, Cu, Hg, Mn, Pb, Sb, Sr, Tl and W did not change with the disease. The best discriminating variables between patients and controls were Cd, Co, Fe, Ni and Si in serum (91.2% of cases correctly classified), and Al, Cd, Co, Fe, Mo and Si in blood (98.2% of cases properly classified).

Fleming J, Spinoulas A, Zheng ML, Cunningham SC, Ginn SL, Mcquilty RC, Rowe PB, Alexander IE. 2005. Partial correction of sensitivity to oxidant stress in

Friedreich ataxia patient fibroblasts by frataxin-encoding adeno-associated virus and lentivirus vectors. Hum Gene Ther 16(8):947-956.
Abstract: Peripheral nervous system (PNS) sensory neurons are directly involved in the pathophysiology of a number of debilitating inherited and acquired neurological conditions. The lack of effective treatments for many such conditions provides a strong rationale for exploring novel therapeutic approaches, including gene therapy. Friedreich ataxia (FRDA), a sensory neuropathy, is a progressive neurodegenerative disease associated with a loss of large sensory neurons from the dorsal root ganglia. Because a mouse model for this well-characterized disease has been generated, we elected to use FRDA as a model disease. In previous studies we achieved efficient and sustained delivery of a reporter gene to PNS sensory neurons, using recombinant adeno-associated viral (AAV) and lentiviral (LV) vectors. In the current study, AAV and LV vectors encoding the human frataxin cDNA were constructed and assessed for frataxin expression and function in primary FRDA patient fibroblast cell lines. FRDA fibroblasts have been shown to exhibit subtle biochemical changes, including increased mitochondrial iron and sensitivity to oxidant stress. Despite the inherent difficulty in working with primary cells, transduction of patient fibroblasts with either vector resulted in the expression of appropriately localized frataxin and partial reversal of phenotype.

Fitsanakis VA, Piccola G, Aschner JL, Aschner M. 2005. Manganese transport by rat brain endothelial (RBE4) cell-based transwell model in the presence of astrocyte conditioned media. J Neurosci Res 81(2):235-243.
Abstract: Manganese (Mn), an essential nutrient, is neurotoxic at high levels and has been associated with the development of a parkinsonian syndrome termed manganism. Currently, the mechanisms responsible for transporting Mn across the blood-brain barrier (BBB) are unknown. By using rat brain endothelial 4 (RBE4) cell monolayers cultured in astrocyte-conditioned media (ACM), we examine the effects of temperature, energy, proton (pH), iron (Fe), and sodium (Na+) dependence on Mn transport. Our results suggest that Mn transport is temperature, energy, and pH dependent, but not Fe or Na+-dependent. These data suggest that Mn transport across the BBB is an active process, but they also demonstrate that the presence of ACM in endothelial cell cultures decreases the permeability of these cells to Mn, reinforcing the use of ACM or astrocyte cocultures in studies examining metal transport across the BBB. (c) 2005 Wiley-Liss, Inc.

Finley BL, Santamaria AB. 2005. Current evidence and research needs regarding the risk of manganese-induced neurological effects in welders. Neurotoxicology 26(2):285-289.

Filipov NM, Seegal RF, Lawrence DA. 2005. Manganese potentiates in vitro production of proinflammatory cytokines and nitric oxide by microglia through a nuclear factor kappa B-dependent mechanism. Toxicol Sci 84(1): 139-148.
Abstract: Recent evidence suggests that the mechanism of manganese (Mn) neurotoxicity involves activation of microglia and/or astrocytes; as a consequence, neurons adjacent to the activated microglia may be injured. Mn modulation of proinflammatory cytokine expression by microglia has not been investigated. Therefore, the objectives of this research were to (1) assess whether Mn induces proinflammatory cytokine expression and/or modulates lipopolysaccharide (LPS)-induced expression of proinflammatory cytokines and (2) investigate possible mechanisms for such an induction. N9 microglia were exposed in vitro to increasing concentrations (50-1000 muM) of Mn in the presence or absence of LPS (10, 100, or 500 ng/ml). After various incubation times (up to 48 h), media levels of several cytokines and nitric oxide (NO) were determined, as was the expression of the inducible form of NO synthase (iNOS). Lactate dehydrogenase (LDH) release into the medium and the cellular uptake of Neutral Red were used as general measures for cytotoxicity. In the absence of LPS, Mn moderately increased interleukin-6 and tumor necrosis factor alpha (TNF-a) production only at higher Mn concentrations, which

were cytotoxic. At all LPS doses, however, proinflammatory cytokine production was dose-dependently increased by Mn. Similarly, LPS-induced NO production and iNOS expression were substantially enhanced by Mn. Pharmacological manipulations indicated that nuclear factor kappa B (NFkappaB) activation is critical for the observed enhancement of cytokine and NO production. Within the context of inflammation, increased production of proinflammatory cytokines and NO by Mn could be an important part of the mechanism by which Mn exerts its neurotoxicity.

Fernaeus S, Land T. 2005. Increased iron-induced oxidative stress and toxicity in scrapie-infected neuroblastoma cells. Neurosci Lett 382(3):217-220.
Abstract: The mechanisms behind the pathology of prion diseases are still unknown, but accumulating evidence suggests oxidative impairment along with metal imbalances in scrapie-infected brains. In this Study, we have investigated iron-induced oxidative stress in scrapie-infected mouse neuroblastoma N2a (ScN2a) cells. Uninfected N2a and ScN2a cells were treated with ferric ammonium citrate (FAC) for 1-16 h, and the levels of labile iron pool (LIP), the formation of reactive oxygen species (ROS), cell viability and ferritin protein levels were measured. The increase in LIP in N2a cells was transient with a quick recovery to normal levels within 4 It accompanied by a moderate increase of formation of ROS after 3 h followed by the decrease to the basal level. In ScN2a cells, the increase in LIP was lower, but the process of recovery was prolonged and accompanied by high ROS formation and decreased cell viability. Ferritin protein levels were significantly lower in ScN2a cells than in wild-type cells in all iron treatments. These results suggest that ScN2a cells are more sensitive to iron treatment as compared to wild-type cells with respect to ROS formation and cell viability, and that ferritin deficiency in infected cells may contribute to iron-induced oxidative stress in scrapie-infected cells. (c) 2005 Elsevier Ireland Ltd. All rights reserved.

Fernaeus S, Halldin J, Bedecs L, Land T. 2005. Changed iron regulation in scrapie-infected neuroblastoma cells. Molecular Brain Research 133(2): 266-273.
Abstract: Prion diseases are characterized by the conversion of the normal cellular prion protein PrPC into a pathogenic isoform, PrPSc. The mechanisms involved in neuronal cell death in prion diseases are largely unknown, but accumulating evidence has demonstrated oxidative impairment along with metal imbalances in scrapie-infected brains. In this study, we report changes in cellular iron metabolism in scrapie-infected mouse neuroblastoma N2a cells (ScN2a). We detected twofold lower total cellular iron and calcein-chelatable cytosolic labile iron pool (LIP) in ScN2a cells as compared to the N2a cells. We also measured in ScN2a cells significantly lower activities of iron regulatory proteins 1 and 2 (IRP1 and IRP2, respectively), regulators of cellular iron by sensing cytosolic free iron levels and controlling posttranscriptionally the expression of the major iron transport protein transferrin receptor 1 (TfR1) and the iron sequestration protein ferritin. IRP1 and IRP2 protein levels were decreased by 40% and 50%, respectively, in ScN2a cells. TfR1 protein levels were fourfold reduced and ferritin levels were threefold reduced in ScN2a cells. TfR1 and ferritin mRNA levels were significantly reduced in ScN2a cells. ScN2a cells responded normally to iron and iron chelator treatment with respect to the activities of IRP1 and IRP2, and biosynthesis of TfR1 and ferritin. However, the activities of IRPI and IRP2, and protein levels of TfR1 and ferritin, were still significantly lower in iron-depleted ScN2a cells as compared to the N2a cells, suggesting lower need for iron in ScN2a cells. Our results demonstrate that scrapie infection leads to changes in cellular iron metabolism, affecting both total cellular and cytosolic free iron, and the activities and expression of major regulators of cellular iron homeostasis. (C) 2004 Elsevier B.V. All rights reserved.

Felix K, Manna SK, Wise K, Barr J, Ramesh GT . 2005. Low levels of arsenite activates nuclear factor-kappa B and activator protein-1 in immortalized mesencephalic cells. J Biochem Mol Toxicol 19(2):67-77.
Abstract: Degeneration of dopaminergic neurons is one of the major

features of Parkinson's disease. Many redox-active metals such as iron and manganese have been implicated in neuronal degeneration characterized by symptoms resembling Parkinson's disease. Even though, arsenic, which is another redox-active metal, has been shown to affect the central monoaminergic systems, but its potential in causing dopaminergic cell degeneration has not been fully known. Hence, the present study was designed to investigate arsenic signaling especially that is mediated by reactive oxygen species and its effect on early transcription factors in dopamine producing mesencephalic cell line 1RB(3)AN(27). These mesencephalic cells were treated with low concentrations of sodium arsenite (0.1, 0.5, 1, 5, and 10 mu M) and incubated for different periods of time (0-4 h). Arsenite was cytotoxic at 5 and 10 mu M concentrations only after 72-h incubation period. Arsenite, in a dose-dependent manner, induced generation of reactive oxygen species (ROS) and activation of early transcription factors such as nuclear factor-kappa B (NF-kappa B) and activator protein-1 (AP-1) as shown by electro mobility shift assay. Incubation of antioxidants, either N-acetyl-L-cysteine (50 mu M) or alpha-tocopherol (50 mu M) with 1 mu M arsenite, suppressed ROS generation. Arsenite at 1 mu M concentration was sufficient for maximal activation of NF-kappa B and AP-1 activation. Time kinetics studies showed maximal activation of NF-kappa B by 1 mu M concentration of arsenite was seen at 120 min and correlated with complete degradation of T kappa B alpha at 60 min. Similarly, maximal activation of AP-1 by 1 mu M concentration of arsenite occurred at 120 min. N-acetyl-L-cysteine at 50 mu M concentration inhibited arsenite-induced NF-kappa B and AP-1. In addition, arsenite was shown to induce phosphorylation of extracellular signal regulated kinase (ERK) 1/2 at concentrations of 1 mu M and above. These results suggest that arsenite, at low and subcytoxic concentrations, appears to induce oxidative stress leading to activation of early transcription factors whereas addition of antioxidant inhibited the activation of these factors. (c) 2005 Wiley Periodicals, Inc.

Fedorow H, Tribl F, Halliday G, Gerlach A, Riederer P, Double KL. 2005. Neuromelanin in human dopamine neurons: Comparison with peripheral melanins and relevance to Parkinson's disease. Prog Neurobiol 75(2): 109-124.

Abstract: Neuromelanin (NM) is a dark polymer pigment produced in specific populations of catecholaminergic neurons in the brain. It appears in greatest quantities in the human brain, in lesser amounts in some other non-human primates, but is absent from the brain in many lower species. Interest in this pigment has seen a resurgence in recent years because of a hypothesised link between neuromelanin and the especial vulnerability of neuromelanin-containing neurons to cell death in Parkinson's disease (PD). Little is known regarding the biology of neuromelanin. As neuromelanin appears to have characteristics in common with the better studied peripheral melanin pigments this review compares what is known about neuromelanin with melanins found in other body tissues. Unlike peripheral melanins, which are produced in specialised cells called melanocytes and may be transferred to other cell types, neuromelanin granules are believed to be stored in the cell in which they are produced. Neuromelanin granules display a unique, more heterogeneous appearance compared with peripheral melanins. Unlike melanin, neuromelanin is traditionally thought to result from a non-enzymatic synthesis pathway with no known pathway for neuromelanin catabolism. More recent data, however, is indicative of some regulation of neuromelanin synthesis and turnover. By analogy with peripheral melanins, neuromelanin may function in vivo to attenuate the effects of damaging stimuli. Among several possible mechanisms suggested, the ability of neuromelanin to interact with transition metals, especially iron, and to mediate intracellular oxidative mechanisms has received particular attention. Recent data from neuromelanin in the Parkinson's disease brain suggests that this proposed function may be compromised, thus rendering pigmented neurons vulnerable to oxidative damage in this disorder. (c) 2005 Elsevier Ltd. All rights reserved.

Erikson KM, Syversen T, Aschner JL, Aschner M. 2005. Interactions between

excessive manganese exposures and dietary iron-deficiency in neurodegeneration. Environ Toxicol Pharmacol 19(3. Sp. Iss. Si):415-421.
Abstract: For nearly a century, manganese has been recognized as an essential nutrient for proper bone formation, lipid, amino acid and carbohydrate metabolism. While manganese deficiency is characterized by symptoms ranging from stunted growth and poor bone remodeling to ataxia, it is manganese toxicity that is far more devastating from a public health standpoint. Most cases of manganese toxicity are the result of occupational exposure to high levels of the metal, and are characterized by specific neurological symptoms referred to as manganism. While manganism shares many common features with Parkinson's disease, there are distinct differences between the two disorders suggesting that manganism might indirectly affect nigrostriatal dopaminergic function. Recent studies from our laboratory show that dietary iron deficiency is a risk factor for brain manganese accumulation and that the striatum is particularly vulnerable. This review briefly discusses manganese from nutritional and toxicological aspects. © 2005 Elsevier B.V. All rights reserved.

Edwin Shackelford R, Manuszak RP, Heard SC, Link CJ, Wang S. 2005. Pharmacological manipulation of ataxia-telangiectasia kinase activity as a treatment for Parkinson's disease. Med Hypotheses 64(4):736-41.
Abstract: Parkinson's disease (PD) is a major cause of morbidity and mortality among older individuals. Although the causes of Parkinson's disease are multifactorial, considerable evidence indicates that elevated labile iron in the substantia nigra pars compacta plays an important role in producing oxyradicals which subsequently damage nigro-striatal neurons. Based on this several researchers have suggested that blood-brain barrier crossing iron chelators might have clinical efficacy in treating PD. Work demonstrating that iron chelators protect nigro-striatal neurons in the N-methyl-4-phenyl-1,2,3,6-tetrahydropyridine and 6-hydroxydopamine-induced rodent PD models supports this hypothesis. Recently, we found that the ATM gene product (mutated in ataxia-telangiectasia, A-T), is required for cell survival and genomic stability maintenance following exposure to low labile iron concentrations. Iron chelators (desferal, quercetin, and apoferritin) also increase A-T cell genomic stability and viability, and activate ATM-dependent cellular events in normal cells. Additionally Atm-deficient mice exhibit a selective loss of dopaminergic nigro-striatal neurons. Based on this, we propose that iron chelators protect the substantia nigra pars compacta not only by chelating labile iron and reducing oxyradical formation, but also by inducing ATM activity, leading to increased oxidative stress resistance and DNA repair. Support for this hypothesis comes from the recent observation that the iron chelating flavonoid quercetin both directly activates ATM and protects neuronal cells from the toxic effects of the N-methyl-4-phenyl-1,2,3,6-tetrahydropyridine. Therefore since; (1) ATM is required for iron toxicity resistance, (2) iron chelators such as quercetin, desferal, and apoferritin induce ATM activity and/or ATM-dependent events, and (3), Atm-deficient mice preferentially lose dopaminergic nigro-striatal neurons, we propose that ATM activity has an important function in PD. Furthermore, pharmacological manipulation of ATM activity via iron chelation might have clinical efficacy in PD treatment.

Ebadi M, Sharma SK, Ghafourifar P, Brown-Borg H, El Refaey H. 2005. Peroxynitrite in the Pathogenesis of Parkinson's Disease and the Neuroprotective Role of MetallothioneinsVolume 396. p 276-+. Nitric Oxide, Pt E: Methods in Enzymology.
Abstract: Parkinson's disease (PD) is characterized by a progressive loss of dopaminergic neurons in the substantia nigra zona compacta and in other subcortical nuclei associated with a widespread occurrence of Lewy bodies. The causes of cell death in Parkinson's disease are still poorly understood, but a defect in mitochondrial oxidative phosphorylation and enhanced oxidative stress has been proposed. We have examined 3-morpholinosydnonimine (SIN-1)-induced apoptosis in control and metallothionein-overexpressing dopaminergic neurons, with a primary

objective to determine the neuroprotective potential of metallothionein (MT) against peroxynitrite-induced neurodegeneration in PD. SIN-1 induced lipid peroxidation and triggered plasma membrane blebbing. In addition, it caused DNA fragmentation, alpha-synuclein induction, and intramitochondrial accumulation of metal ions (copper, iron, zinc, and calcium), and it enhanced the synthesis of 8-hydroxy-2-deoxyguanosine. Furthermore, it downregulated the expression of Bcl-2 and poly(adenosine diphosphate-ribose) polymerase, but upregulated the expression of caspase-3 and Bax in dopaminergic (SK-N-SH) neurons. SIN-1 induced apoptosis in aging mitochondrial genome knockout cells, alpha-synuclein-transfected cells, metallothionein double-knockout cells, and caspase-3-overexpressed dopaminergic neurons. SIN-1-induced changes were attenuated with selegiline or in metallothionein-transgenic striatal fetal stem cells. SIN-1-induced oxidation of dopamine (DA) to dihydroxyphenylacetaldehyde (DopaL) was attenuated in metallothionein-transgenic fetal stem cells and in cells transfected with a mitochondrial genome, and was enhanced in aging mitochondrial genome knockout cells, in metallothionein double-knockout cells, and caspase-3 gene-overexpressing dopaminergic neurons. Selegiline, melatonin, ubiquinone, and metallothionein suppressed SIN-1-induced downregulation of a mitochondrial genome and upregulation of caspase-3 as determined by reverse transcription polymerase chain reaction. These studies provide evidence that nitric oxide synthase activation and peroxynitrite ion overproduction may be involved in the etiopathogenesis of PD, and that metallothionein gene induction may provide neuroprotection.

Ebadi M, Brown-Borg H, El Refaey H, Singh BB, Garrett S, Shavali S, Sharma SK. 2005. Metallothionein-mediated neuroprotection in genetically engineered mouse models of Parkinson's disease. Molecular Brain Research 134(1): 67-75.

Abstract: Parkinson's disease is characterized by a progressive loss of dopaminergic neurons in the substantia nigra zona compacta, and in other sub-cortical nuclei associated with a widespread occurrence of Lewy bodies. The cause of cell death in Parkinson's disease is still poorly understood, but a defect in mitochondrial oxidative phosphorylation and enhanced oxidative and nitrative stresses have been proposed. We have studied control(wt) (C57B1/6), metallothionein transgenic (MTtrans), metallothionein double gene knock (MTdko), alpha-synuclein knock out (alpha-syn(ko)), alpha-synuclein-metallothionein triple knock out (alpha-syn-MTtko), weaver mutant (wv/wv) mice, and Ames dwarf mice to examine the role of peroxynitrite in the etiopathogenesis of Parkinson's disease and aging. Although MTdko mice were genetically susceptible to 1, methyl, 4-phenyl, 1,2,3,6-tetrahydropyridine (MPTP) Parkinsonism, they did not exhibit any overt clinical symptoms of neurodegeneration and gross neuropathological changes as observed in wv/wv mice. Progressive neurodegenerative changes were associated with typical Parkinsonism in wv/wv mice. Neurodegenerative changes in wv/wv mice were observed primarily in the striatum, hippocampus and cerebellum. Various hallmarks of apoptosis including caspase-3, TNF alpha, NF kappa B, metallothioneins (MT-1, 2) and complex-1 nitration were increased; whereas glutathione, complex-1, ATP, and Ser(40)-phosphorylation of tyrosine hydroxylase, and striatal F-18-DOPA uptake were reduced in wv/wv mice as compared to other experimental genotypes. Striatal neurons of wv/wv mice exhibited age-dependent increase in fibs, c-jun, caspase-3, and GAPDH) induction, interdense cored intra-neuronal inclusions, cellular aggregation, proto-oncogenes (c-fos, c-jun,caspase-3, and GAPDH) introduction, nucleosomal DNA fragmentation, and neuro-apoptosis. MTtrans and alpha-Syn(ko) mice were genetically resistant to MPTP-Parkinsonism and Ames dwarf mice possessed significantly higher concentrations of striatal coenzyme Q(10) and metallothioneins (MT 1, 2) and lived almost 2.5 times longer as compared to control(wt) mice. A potent peroxynitrite ion generator, 3-morpholinosydnonimine (SIN-1)-induced apoptosis was significantly attenuated in MTtrans fetal stem cells. These data are interpreted to suggest that peroxynitrite ions are involved in the etiopathogenesis of Parkinson's disease, and metallothionein-mediated coenzyme Q(10)

synthesis may provide neuroprotection. (C) 2004 Elsevier B.V. All rights reserved.

Eakin CM, Miranker AD. 2005. From chance to frequent encounters: Origins of beta 2-microglobulin fibrillogenesis. Biochimica Et Biophysica Acta-Proteins and Proteomics 1753(1):92-99.
Abstract: It is generally accepted that amyloid formation requires partial, but not complete unfolding of a polypeptide chain. Amyloid formation by beta-2 microglobulin (beta 2m), however, readily occurs under strongly native conditions provided that there is exposure to specific transition metal cations. In this review, we discuss transition metal catalyzed conformational changes in several amyloidogenic systems including prion protein, Alzheimer's and Parkinson's diseases. For some systems, including beta 2m from dialysis related amyloidosis (DRA), catalysis overcomes an entropic barrier to protein aggregation. Recent data suggest that beta 2m samples conformations that are under thermodynamic control, resulting in local or partial unfolding under native conditions. Furthermore, exposure to transition metal cations stabilizes these partially unfolded states and promotes the formation of small oligomers, whose structures are simultaneously near-native and amyloid-like. By serving as a tether, Cu2+ enables the encounter of amyloidogenic conformations to occur on time scales which are significantly more rapid than would occur between freely diffusing monomeric protein. Once amyloid formation occurs, the requirement for Cu2+ is lost. We assert that beta 2m amyloid fiber formation at neutral pH may be facilitated by rearrangements catalyzed by the transient and pair wise tethering of beta 2m at the blood/dialysate interface present during therapeutic hemodialysis. (c) 2005 Published by Elsevier B.V.

Dodd CA, Ward DL, Klein BG. 2005. Basal ganglia accumulation and motor assessment following manganese chloride exposure in the C57BL/6 mouse. International Journal of Toxicology 24(6):389-397.
Abstract: Equivocal clinical evidence for involvement of manganese in development of Parkinson's disease necessitates experimental studies on this issue. The aged, 1-methyl-4-phenyl-1,2,3,6-tetrahyropyridine-treated C57BL/6 mouse is one of the most common models for Parkinson's disease. However, there is little information on brain bioaccumulation of manganese, and little or no information on clinical/behavioral manifestations of manganese neurotoxicity, in this strain. Male C57BL/6 retired breeder mice were given a single subcutaneous injection of either 0, 50, or 100 mg/kg of MnCl2 (single-dose regimen) or three injections of either of these doses over 7 days (multiple-dose regimen). Behavioral assessment was performed 24 h after final injection, followed by sacrifice, and body weight was recorded each day. There was a 105% increase in striatal manganese concentration 1 day after a single 100 mg/kg injection, and 421% and 647% increases, respectively, 1 day after multiple doses of 50 or 100 mg/kg of MnCl2. One day after a single injection, there were respective 30.9% and 38.9% decreases in horizontal movement (grid crossing) for the 50 and 100 mg/kg doses and a 43.2% decrease for the multiple dose of 100 mg/kg. There was no significant main effect of dose level on rearing, swimming, grip strength, or grip fatigue. Unlike previous work with the C57BL/6 strain using smaller intraperitoneal doses, this study established dosing regimens that produced significant increases in basal ganglia manganese concentration reminiscent of brain increases in the CD-1 mouse following subcutaneous doses close to our lowest. A decrease in locomotor behavior, significant but not severe in this study, has been reported following manganese exposure in other mouse strains. These data, particularly the significant increase in basal ganglia manganese concentration, provide guidance for designing studies of the potential role of manganese in Parkinson's disease using the most common animal model for the disorder.

Dieter HH, Bayer TA, Multhaup G. 2005. Environmental copper and manganese in the pathophysiology of neurologic diseases (Alzheimer's disease and manganism). Acta Hydrochimica Et Hydrobiologica 33(1):72-78.

Abstract: Alzheimer's disease (AD) is the most common form of dementia. Populations migrating from developing to industrialized countries seem to elicit a higher incidence and prevalence rate of AD, suggesting lifestyle and environmental factors to have a role in the Pathophysiology of AD. One of its major neuropathological hallmarks is the deposition of AB peptides as amyloid plaques in the brain of AD patients. A beta is proteolytically cleaved out of the larger amyloid precursor protein (APP). Cu and Mn are often found in drinking water and may have a neurotoxic potential. APP is involved in Cu homeostasis in mouse and man. In vitro observations and in vivo data obtained from APP mouse models provide strong evidence that APP overexpression enables intracellular Cu to be transported out of the cell. Disturbed metal-ion homeostasis with elevated serum Cu levels occurs in Alzheimer and Down's patients and lowered levels in post-mortem AD brain. We observed that bioavailable Cu has specific beneficial effects in an Alzheimer's disease mouse model. This should be regarded as a proof-of-concept for a. prophylactic approach to overcome the observed CNS Cu deficiency in the brain of Alzheimer's disease patients. Manganism is a disorder with symptoms similar to that of Parkinson's disease. The precise mechanism how manganese can damage the nervous system is unknown. There is some evidence that iron and manganese may utilize similar transport systems. Epidemiologic data strongly suggests that manganese enters the body primarily via inhalation and through the ingestion of manganese in drinking water.

De Lima MNM, Polydoro M, Laranja DC, Bonatto F, Bromberg E, Moreira JCF, Dal-Pizzol F, Schroder N. 2005. Recognition memory impairment and brain oxidative stress induced by postnatal iron administration. Eur J Neurosci 21 (9):2521-2528.
Abstract: Iron accumulation in the brain has been implicated in the pathogenesis of neurodegenerative disorders. It is known that iron catalyses the formation of highly reactive hydroxyl radicals. Recent studies have implicated oxidative damage in memory deficits in rats and humans. The purpose of the present study was to investigate the long-term effects of iron treatment in four different phases of the neonatal period on recognition memory in rats. Additionally, parameters of oxidative stress in cerebral regions related to memory formation were evaluated. Male Wistar rats received vehicle or 10.0 mg/kg of Fe2+ orally at postnatal days 5-7, 12-14, 19-21 or 30-32. Animals given iron at any phase of the neonatal period showed impairments in long-term retention of object recognition memory, although only the group given iron from postnatal days 12-14 showed a complete memory blockade. Iron treatment induced oxidative damage in the brain as assessed by the thiobarbituric acid reactive species assay. Moreover, iron administration increased superoxide production in submitochondrial particles, suggesting impaired mitochondrial function; and there was an increase in superoxide dismutase activity in brain regions susceptible to iron administration. The results show that iron load in the early stages of life induces cognitive impairment possibly by inducing oxidative damage in the brain. These findings are consistent with the view that oxidative stress may be related to the cognitive decline observed in normal ageing.

De Lima MNM, Laranja DC, Caldana F, Grazziotin MM, Garcia VA, Dal-Pizzol F, Bromberg E, Schroder N. 2005. Selegiline protects against recognition memory impairment induced by neonatal iron treatment. Exp Neurol 196 (1):177-183.
Abstract: Excess of iron in the brain has been implicated in the pathogenesis of several human neurodegenerative diseases, for example Alzheimer's disease and Parkinson's disease. It has been shown that the neonatal period is critical for the establishment of normal iron content in the adult brain. Moreover, it is known that aging alters the cerebral distribution of this metal. We have recently described that neonatal administration of iron severely impaired novel object recognition memory in rats. The aim of the present study was to determine whether selegiline, a monoamine oxidase (MAO) inhibitor known for its neuroprotective properties, could protect rats against cognitive impairment induced by

neonatal administration of iron. In the first experiment, male Wistar rats received vehicle (5% sorbitol in water) or iron (10.0 mg/kg) orally from postnatal days 12 to 14 and saline (0.9% NaCl) or selegiline (1.0 or 10.0 mg/kg) intraperitoneally for 21 days, starting 24 h before the first iron dosing. In the second experiment, rats were given either vehicle or iron (10.0 mg/kg) orally from postnatal days 12 to 14 followed by saline or selegiline (1.0 or 10.0 mg/kg) intraperitoneally for 21 days, starting when rats reached adulthood (50th day after birth). Iron-treated rats given selegiline in both doses showed no deficits in recognition memory. Rats receiving iron but no selegiline presented memory deficits. This is the first study reporting the reversion of iron-induced memory impairment, supporting the view that our model can be considered as a useful tool in the search for new drugs With neuroprotective and/or memory enhancing properties. (c) 2005 Elsevier Inc. All rights reserved.

Dalfo E, Portero-Otin M, Ayala V, Martinez A, Pamplona R, Ferrer I. 2005. Evidence of oxidative stress in the neocortex in incidental Lewy body disease. J Neuropathol Exp Neurol 64(9):816-830.
Abstract: Oxidative stress has been well documented in the substantia nigra in Parkinson disease (PD), but little is known about oxidative damage, particularly lipoxidation, advanced glycation (AGE), and AGE receptors (RAGE) in other structures, including the cerebral cortex, in early stages of diseases with Lewy bodies. The present study was undertaken to analyze these parameters in the frontal cortex (area 8), amygdala, and substantia nigra in selected cases with no neurologic symptoms and with neuropathologically verified incidental Lewy body disease-related changes, comparing them with healthy age-matched individuals. Results of the present study have shown mass spectrometric and immunologic evidences of increased lipoxidative damage by the markers malondialdehyde-lysine (MDAL) and 4-hydroxynonenallysine (HNE), increased expression of AGE in the substantia nigra, amygdala, and frontal cortex, and increased and heterogeneous RAGE cellular expression in the substantia nigra and frontal cortex in cases with early stages of parkinsonian neuropathology. In addition, increased content of the highly peroxidizable docosahexaenoic acid in the amygdala and frontal cortex. These changes were not associated to a-synuclein aggregation in cortex, contrasting with aggregates found in SDS-soluble fractions of frontal cortex in dementia with Lewy bodies (DLB) cases. The pattern of lipidic abnormalities differed in DLB and incidental Lewy body disease. Furthermore, although AGE and RAGE expression were raised in DLB, no increase in the total amount of HNE and MDAL adducts was found in the cerebral cortex in DLB. Preliminary analyses have identified 2 proteins with lipoxidative damage, alpha-synuclein and manganese superoxide dismutase (SOD2), in incidentally Lewy body disease cortex. This study demonstrates abnormal fatty acid profiles, increased and selective lipoxidative damage, and increased AGE and RAGE expression in the frontal cortex in cases with early stages of parkinsonian neuropathology without treatment. These findings further support antioxidant therapy in the treatment of PD to reduce cortical damage associated with oxidative stress.

Cui ZR, Lockman PR, Atwood CS, Hsu CH, Gupte A, Allen DD, Mumper RJ. 2005. Novel D-penicillamine carrying nanoparticles for metal chelation therapy in Alzheimer's and other CNS diseases. Eur J Pharm Biopharm 59(2): 263-272.
Abstract: Metal ions accumulate in the brain with aging and in several neurodegenerative diseases. Aside from the copper storage disease, Wilson's disease, recent attention has focused on the accumulation of zinc, copper and iron in the Alzheimer's disease (AD) brain and the accumulation of iron in Parkinson's disease. In particular, the parenchymal deposition of beta-amyloid (Abeta) and its interaction with metal ions has been postulated to play a role in the progression of AD. Thus, the strategy of lowering brain metal ions and targeting the interaction of Abeta peptide and metal ions through the administration of chelators has merit. Our recent finding that nanoparticle delivery systems can cross the blood-brain barrier has led us to investigate whether chelators delivered conjugated to

nanoparticles could act to reverse metal ion induced protein precipitation. In the present studies, the Cu (I) chelator D-penicillamine was covalently conjugated to nanoparticles via a disulfide bond or a thioether bond. Nanoparticle-chelator conjugates were stable between pH 6-8 in aqueous suspension if stored at 4 degreesC, and did not aggregate when challenged with salts and serum. Release Of D-penicillamine from the nanoparicles was achieved using reducing agents such as dithiothreitol (as a model for glutathione). Nanoparticles treated only under reducing conditions that released the conjugated D-penicillamine were able to effectively resolubilize copper-Abeta (1-42) aggregates. These results indicate that nanoparticles have potential to deliver D-penicillamine to the brain for the prevention of AD (1-42) accumulation, as well as to reduce metal ion accumulation in other CNS diseases. (C) 2004 Elsevier B.V. All rights reserved.

Crossgrove JS, Yokel RA. 2005. Manganese distribution across the blood-brain barrier - IV. Evidence for brain influx through store-operated calcium channels. Neurotoxicology 26(3):297-307.
Abstract: Manganese (Mn) is a required co-factor for many ubiquitous enzymes; however chronic Mn soverexposure can cause manganisin, a parkinsonian-like syndrome. Previous studies showed Mn influx into brain is carrier-mediated, though the Initiative carrier(s) ivere not established. Studies conducted with cultured bovine brain microvascular endothelial cells (bBMECs), which comprise the blood-brain barrier, revealed Mn-54 (H) uptake positivel'y correlated with pH, was temperature-dependent, and was sodium-and energy-independent. Brain Mn-54 uptake correlated inversely with calcium (Ca) concentration, but Ca-45 uptake was unaltered by high Mn concentration. Lanthanum (La), a non-selective inhibitor of several Ca channel types, as well as verapainil and amiloride, inhibitors of voltage-operated Ca channels,fiailed to inhibit Mn uptake into cells. Nickel (Ni), another non-selective inhibitor of several Ca channel types, inhibited Mn and Ca uptake into cells by 88 and 85%, respectively. Cyclopiazonic acid (CPA) and thapsigargin, which activate store-operated calcium channels (SOCCs), increased Mn-54 and Ca-45 uptake into cultured bBMECs. In situ brain perfusion studies were conducted ill adult, male Sprague-Dawley rats to verify; the cell culture results. Both nickel and verapamil produced a non-significant decrease in Mn and Ca influx. Lanthanion significantly increased Mn influx to 675 and 450% of control ill parietal cortex and caudate, respectively, while producing no significant effect oil Ca influx. Vanadote, which inhibits Ca-ATPase, inhibited Mn uptake into cultured blood-brain barrier cells, but not into perfused rat brain. Overall these results suggest that both Ca-dependent and Ca-independent mechanisms play a role in brain Mn influx. This work provides evidence that store-operated Ca channels, as well as another mechanism at the blood-brain barrier, likely play a role tit carrier-mediated Mn influx into the brain. (c) 2004 Elsevier Inc. All rights reserved.

Collingwood JF, Mikhaylova A, Davidson M, Batich C, Streit WJ, Terry J, Dobson J. 2005 . In situ characterization and mapping of iron compounds in Alzheimer's disease tissue. Journal of Alzheimers Disease 7(4):267-272.
Abstract: There is a well-established link between iron overload in the brain and pathology associated with neurodegeneration in a variety of disorders such as Alzheimer's (AD), Parkinson's (PD) and Huntington's (HD) diseases [1]. This association was first discovered in AD by Goodman in 1953 [2], where, in addition to abnormally high concentrations of iron in autopsy brain tissue, iron has also been shown to accumulate at sites of brain pathology such as senile plaques [3]. However, since this discovery, progress in understanding the origin, role and nature of iron compounds associated with neurodegeneration has been slow. Here we report, for the first time, the location and characterisation of iron compounds in human AD brain tissue sections. Iron fluorescence was mapped over a frontal-lobe tissue section from an Alzheimer's patient, and anomalous iron concentrations were identified using synchrotron X-ray absorption techniques at 5 mu m spatial resolution. Concentrations of ferritin and magnetite, a magnetic iron oxide potentially indicating disrupted brain-iron

metabolism, were evident. These results demonstrate a practical means of correlating iron compounds and disease pathology in-situ and have clear implications for disease pathogenesis and potential therapies.

Cole NB, Murphy DD, Lebowitz J, Di Noto L, Levine RL, Nussbaum RL. 2005. Metal-catalyzed oxidation of alpha-synuclein - Helping to define the relationship between oligomers, protofibrils, and filaments. J Biol Chem 280(10): 9678-9690.
Abstract: Oxidative stress is implicated in a number of neuro-degenerative diseases and is associated with the selective loss of dopaminergic neurons of the substantia nigra in Parkinson's disease. The role of alpha-synuclein as a potential target of intracellular oxidants has been demonstrated by the identification of posttranslational modifications of synuclein within intracellular aggregates that accumulate in Parkinson's disease brains, as well as the ability of a number of oxidative insults to induce synuclein oligomerization. The relationship between these relatively small soluble oligomers, potentially neurotoxic synuclein protofibrils, and synuclein filaments remains unclear. We have found that metal-catalyzed oxidation of alpha-synuclein inhibited formation of synuclein filaments with a concomitant accumulation of beta sheet-rich oligomers that may represent synuclein protofibrils. Similar results with a number of oxidative and enzymatic treatments suggest that the covalent association of synuclein into higher molecular mass oligomers/ protofibrils represents an alternate pathway from filament formation and renders synuclein less prone to proteasomal degradation.

Chwiej J, Szczerbowska-Boruchowska M, Lankosz M, Wojcik S, Falkenberg G, Stegowski Z, Setkowicz Z. 2005. Preparation of tissue samples for X-ray fluorescence microscopy. Spectrochimica Acta Part B-Atomic Spectroscopy 60(12):1531-1537.
Abstract: As is well-known, trace elements, especially metals, play an important role in the pathogenesis of many disorders. The topographic and quantitative elemental analysis of pathologically changed tissues may shed some new light on processes leading to the degeneration of cells in the case of selected diseases. An ideal and powerful tool for such purpose is the Synchrotron Microbeam X-ray Fluorescence technique. It enables the carrying out of investigations of the elemental composition of tissues even at the single cell level. The tissue samples for histopathological investigations are routinely fixed and embedded in paraffin. The authors try to verify the usefulness of such prepared tissue sections for elemental analysis with the use of X-ray fluorescence microscopy. Studies were performed on rat brain samples. Changes in elemental composition caused by fixation in formalin or paraformaldehyde and embedding in paraffin were examined. Measurements were carried out at the bending magnet beamline L of the Hamburger Synchrotronstrahlungslabor HASYLAB in Hamburg. The decrease in mass per unit area of K, Br and the increase in P, S, Fe, Cu and Zn in the tissue were observed as a result of the fixation. For the samples embedded in paraffin, a lower level of most elements was observed. Additionally, for these samples, changes in the composition of some elements were not uniform for different analyzed areas of rat brain. (c) 2005 Elsevier B.V. All rights reserved.

Chwiej J, Fik-Mazgaj K, Szczerbowska-Boruchowska M, Lankosz M, Ostachowicz J, Adamek D, Simionovici A, Bohic S. 2005. Classification of nerve cells from substantia nigra of patients with Parkinson's disease and amyotrophic lateral sclerosis with the use of X-ray fluorescence microscopy and multivariate methods. Anal Chem 77(9):2895-2900.
Abstract: The causes of Parkinson's disease and amyotrophic lateral sclerosis are still not known, but there is evidence that metal ions can be involved in processes leading to degeneration and atrophy of neurons in the case of these two neurodegenerative disorders. A synchrotron microbeam X-ray fluorescence technique was applied for topographic and quantitative analyses of selected elements on central nervous system tissue. The thin slices of brain were measured on the undulator beamline ID 22 at the European Synchrotron Radiation Facility in Grenoble, France.

The polychromatic beam with the dimension of 5 μ m x 2 μ m (horizontal x vertical) was used in measurements. Tissues of substantia nigra representing Parkinson's disease, amyotrophic lateral sclerosis, and the control case were scanned. The results obtained indicated that accumulation of some elements depends on the case that the substantia nigra represents. Some variability in the elemental distribution for a given case was noticed as well. To investigate if present differences in the elemental accumulation between analyzed cases are statistically significant, multivariate methods were used. Cluster and discriminant analyses confirmed the significance of the differences in elemental accumulation in biological structures representing the examined cases. The methods used let us classify these structures in separate groups and determine elements, which play the greatest role in the differentiation of the biological structures for each case.

Choi JY, Jang EH, Park CS, Kang JH. 2005. Enhanced susceptibility to 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine neurotoxicity in high-fat diet-induced obesity. Free Radic Biol Med 38(6):806-816.
Abstract: Currently, obesity is considered a systemic inflammation; however, the effects of obesity on the vulnerability of dopaminergic neurons to oxidative stress are not fully defined. We evaluated the effects of high-fat diet-induced obesity (HF DIO) on neurotoxicity in mice treated with 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP). Eight weeks after a HF or matched normal diet, a severe decrease in the levels of striatal dopamine and of nigral microtubule-associated protein 2, manganese superoxide dismutase, and tyrosine hydroxylase was observed in obese mice treated with subtoxic doses of MPTP (20 mg/kg) compared with the matched lean group. In addition, the levels of nitrate/nitrite and thiobarbituric acid-malondialdehyde adducts in the substantia nigra of obese mice were reciprocally elevated or suppressed by MPTP. Interestingly, striatal nNOS phosphorylation and dopamine turnover were elevated in obese mice after MPTP treatment, but were not observed in lean mice. The nitrotyrosine immunoreactivity for evaluation of nigral nitrogenous stress in obese mice with MPTP was higher than that in matched lean mice. At higher doses of MPTP (60 mg/kg), the mortality was higher in obese mice than in lean mice. These results suggest that DIO may increase the vulnerability of dopaminergic neurons to MPTP via increased levels of reactive oxygen and nitrogen species, and the role of nNOS phosphorylation in the MPTP toxicities and dopamine homeostasis should be further evaluated. (C) 2004 Elsevier Inc. All rights reserved.

Chinta SJ, Andersen JK. 2005. Dopaminergic neurons. International Journal of Biochemistry & Cell Biology 37(5):942-946.
Abstract: Dopaminergic neurons of the midbrain are the main source of dopamine (DA) in the mammalian central nervous system. Their loss is associated with one of the most prominent human neurological disorders, Parkinson's disease (PD). Dopaminergic neurons are found in a 'harsh' region of the brain, the substantia nigra pars compacta, which is DA-rich and contains both redox available neuromelanin and a high iron content. Although their numbers are few, these dopaminergic neurons play an important role in the control of multiple brain functions including voluntary movement and a broad array of behavioral processes such as mood, reward, addiction, and stress. Studies into the developmental pathways which are involved in the generation of dopaminergic neurons in the brain have led to the identification of several specific transcription factors including Nurr1, Lmx1b and Pitx3, all shown to be important in the development of the mesencephalic dopaminergic system. The selective degeneration of these dopaminergic neurons in the substantia nigra pars compacta leads to PD but the exact cause for this nigral cell loss is still unknown. (c) 2004 Elsevier Ltd. All rights reserved.

Chillrud SN, Grass D, Ross JM, Coulibaly D, Slavkovich V, Epstein D, Sax SN, Pederson D, Johnson D, Spengler JD, Kinney PL, Simpson HJ, Brandt-Rauf P. 2005. Steel dust in the New York City subway system as a source of manganese, chromium, and iron exposures for transit workers. Journal of

Urban Health-Bulletin of the New York Academy of Medicine 82(1):33-42.
Abstract: The United States Clean Air Act Amendments of 1990 reflected increasing concern about potential effects of low-level airborne metal exposure on a wide array of illnesses. Here we summarize results demonstrating that the New York City (NYC) subway system provides an important microenvironment for metal exposures for NYC commuters and subway workers and also describe an ongoing pilot study of NYC transit workers' exposure to steel dust. Results from the TEACH (Toxic Exposure Assessment, a Columbia and Harvard) study in 1999 of 41 high-school students strongly suggest that elevated levels of iron, manganese, and chromium in personal air samples were due to exposure to steel dust in the NYC subway. Airborne concentrations of these three metals associated with fine particulate matter were observed to be more than 100 times greater in the subway environment than in home indoor or outdoor settings in NYC. While there are currently no known health effects at the airborne levels observed in the subway system, the primary aim of the ongoing pilot study is to ascertain whether the levels of these metals in the subway air affect concentrations of these metals or related metabolites in the blood or urine of exposed transit workers, who due to their job activities could plausibly have appreciably higher exposures than typical commuters. The study design involves recruitment of 40 transit workers representing a large range in expected exposures to steel dust, the collection of personal air samples of fine particulate matter, and the collection of blood and urine samples from each monitored transit worker.

Chen YR, Chen CL, Zhang LW, Green-Church KB, Zweier JL. 2005. Superoxide generation from mitochondrial NADH dehydrogenase induces self-inactivation with specific protein radical formation. J Biol Chem 280(45): 37339-37348.
Abstract: Mitochondrial superoxide (O-2(radical anion).) production is an important mediator of oxidative cellular injury. While NADH dehydrogenase (NDH) is a critical site of this O-2(radical anion) production; its mechanism of O-2(radical anion) generation is not known. Therefore, the catalytic function of NDH in the mediation of O-2(radical anion) generation was investigated by EPR spin-trapping. In the presence of NADH, O-2(radical anion) generation from NDH was observed and was inhibited by diphenyleneiodinium chloride (DPI), indicating involvement of the FMN-binding site of NDH. Addition of FMN increased O-2(radical anion) production. Destruction of the cysteine ligands of iron-sulfur clusters decreased O-2(radical anion) generation, suggesting a secondary role of this site. This inhibitory effect was reversed by addition of FMN. However, FMN addition could not reverse the inhibition of NDH by either DPI or heat denaturation, demonstrating involvement of both FMN and its FMN-binding protein moiety in the catalysis of O-2(radical anion) generation. O-2(radical anion) production by NDH also induced self-inactivation. Immunospin-trapping with anti-DMPO antibody and subsequent mass spectrometry was used to define the sites of oxidative damage of NDH. A DMPO adduct was detected on the 51-kDa subunit and was O-2(radical anion)-dependent. Alkylation of the cysteine residues of NDH significantly inhibited NDH-DMPO spin adduct formation, indicating involvement of protein thiyl radicals. LC/MS/MS analysis of a tryptic digest of the 51-kDa polypeptide revealed that cysteine (Cys(206)) and tyrosine (Tyr(177)) were specific sites of NDH-derived protein radical formation. Thus, two domains of the 51-kDa subunit, Gly(200-) Ala-Gly-Ala-Tyr-Ile-Cys(206)- Gly-Glu-Glu- Thr-Ala-Leu-Ile-Glu-Ser-Ile-Glu-Gly-Lys(219) and Ala(176)-Tyr(177)- Glu-Ala-Gly-Leu-Ile-Gly-Lys(184), were demonstrated to be susceptible to oxidative attack, and their oxidative modification results in decreased electron transfer activity.

Chen J, Small-Howard A, Yin A, Berry MJ. 2005. The responses of Ht22 cells to oxidative stress induced by buthionine sulfoximine (BSO). BMC Neuroscience 6(1):10.
Abstract: Background: glutathione (GSH) is the most abundant thiol antioxidant in mammalian cells. It directly reacts with reactive oxygen species (ROS), functions as a cofactor of antioxidant enzymes, and

maintains thiol redox potential in cells. GSH depletion has been implicated in the pathogenesis of neurological diseases, particularly to Parkinson's disease (PD). The purpose of this study was to investigate the change of cellular antioxidant status and basic cell functions in the relatively early stages of GSH depletion. Results: in this study, GSH was depleted by inhibition of glutamylcysteine synthetase using buthionine sulfoximine (BSO) treatment in Ht22, a neuronal cell line derived from mouse hippocampus. Treatment with BSO produced dose-dependent decreases in total GSH level, Fe3+-reducing ability (FRAP assay), Cu2+- reducing ability (Antioxidant Potential, AOP assay), and ABTS free radical scavenging ability (ABTS assay) of the cells, but the sensitivity of these indicators to dosage varied considerably. Most of the changes were completed during the first 8 hours of treatment. Cell viability was tested by MTT (3-[4,5-dimethylthiazol-2-yl]-2,5-diphenyl tetrazolium bromid) assay, and cells at lower density in culture were found to be more sensitive to GSH depletion. The activity of antioxidant enzymes, such as glutathione peroxidase (GPx), glutathione reductase (GR), and copper/zinc superoxide dismutase (Cu/Zn-SOD) were affected by GSH depletion. A cDNA expression array assay of the effects of BSO treatment showed significantly decreased mRNA level for 3 genes, and significantly increased mRNA level for 10 genes, including the antioxidant enzymes Cu/Zn-SOD and thioredoxin peroxidase 2 (TPxII). Conclusions: the study suggests that there are BSO-sensitive and BSO-resistant pools of GSH in Ht22 cells, and that different categories of antioxidant react differently to GSH depletion. Further, the effect of GSH status on cell viability is cell density dependent. Finally, the alterations in expression or activity of several antioxidant enzymes provide insight into the various cellular responses to GSH depletion.

Chang YZ, Qian ZM, Wang K, Zhu L, Yang XD, Du JR, Jiang L, Ho KP, Wang Q, Ke Y. 2005. Effects of development and iron status on ceruloplasmin expression in rat brain. J Cell Physiol 204(2):623-631.
Abstract: The increased iron content in the brain of subjects with aceruloplasminemia has implicated ceruloplasmin (CP) as a major factor in the regulation of regional brain iron content. In this study, we investigated the effects of age and iron on CP expression in rat brain. In all four regions, the iron concentrations increased with developmental age. There is a similar trend in age-induced changes in CP mRNA and protein. The CP mRNA and protein levels were both lowest at postnatal day (PND) 7. The expression increased gradually with age, reaching the highest at PND196 in the striatum and substantia nigra, and at PND21 and PND63 in the cortex and hippocampus, respectively. This suggests the existence of an age-dependent pre-transcriptional regulation and a regionally specific effect of age on CP expression in the brain. Although total iron in all four regions was significantly lower in the rats fed with a low-iron diet for 6 weeks and higher in the rats with a high-iron diet than those in the control animals, no significant between-group differences in CP mRNA and protein were found in these animals, except in the substantia nigra where a significant increase in CP protein in high-iron rats was observed, and the reverse in low-iron rats. These findings suggested that the effects of iron on CP expression in the brain may be region-specific, and that regulation of CP expression by iron in the substantia nigra was at the post-transcriptional level. J. Cell. Physiol. 204: 623-631, 2005. (c) 2005 Wiley-Liss, Inc.

Chan KH, Cheung RTF, Au-Yeung KM, Mak W, Cheng TS, Ho SL. 2005. Wilson's disease with depression and parkinsonism. Journal of Clinical Neuroscience 12(3):303-305.
Abstract: Wilson's disease (WD) is an autosomal recessive disorder with reduced biliary excretion of copper plus impaired formation of ceruloplasmin, leading to copper accumulation in the liver, brain, kidney, and cornea. Clinical manifestations include liver damage, psychiatric symptoms, and neurological features. We report a 35-year-old woman with a history of deranged liver functions who had severe depression several years later and eventually presented with parkinsonian features. The underlying diagnosis is WD and family screening revealed WD in 2 other siblings. She could not tolerate penicillamine because of fever and

leucopenia. While taking trientine hydrochloride and zinc sulphate, her parkinsonism improved and her depression remained in remission. WD should be considered in patients with unexplained liver function derangement or psychiatric symptoms. Early diagnosis and initiation of specific treatment are crucial in minimising any further cerebral and hepatic damage as well as securing possible improvement in organ functions. (c) 2004 Published by Elsevier Ltd.

Cardozo-Pelaez F, Cox DP, Bolin C. 2005. Lack of the DNA repair enzyme OGG1 sensitizes dopamine neurons to manganese toxicity during development. Gene Expr 12(4-6):315-323.
Abstract: Onset of Parkinson's disease (PD) and Parkinson-like syndromes has been associated with exposure to diverse environmental stimuli. Epidemiological studies have demonstrated that exposure to elevated levels of manganese produces neuropathological changes localized to the basal ganglia, including neuronal loss and depletions in striatal dopamine content. However, understanding the mechanisms associated with manganese neurotoxicity has been hampered by the lack of a good rodent model. Elevated levels of 8-hydroxy-2'-deoxyguanosine (oxo(8)dG) have been found in brain areas affected in PD. Whether increased DNA damage is responsible for neuronal degeneration or is a mere epiphenomena of neuronal loss remains to be elucidated. Thus, by using mice deficient in the ability to remove oxo(8)dG we aimed to determine if dysregulation of DNA repair coupled to manganese exposure would be detrimental to dopaminergic neurons. Wild-type and OGG1 knockout mice were exposed to manganese from conception to postnatal day 30; in both groups, exposure to manganese led to alterations in the neurochemistry of the nigrostriatal system. After exposure, dopamine levels were elevated in the caudate of wild-type mice. Dopamine was reduced in the caudate of OGG1 knockout mice, a loss that was paralleled by an increase in the dopamine index of turnover. In addition, the reduction of dopamine in caudate putamen correlated with the accumulation of oxo(8)dG in midbrain. We conclude that OGG1 function is essential in maintaining neuronal stability during development and identify DNA damage as a common pathway in neuronal loss after a toxicological challenge.

Cappai R, Leck SL, Tew DJ, Williamson NA, Smith DP, Galatis D, Sharples RA, Curtain CC, Ali FE, Cherny RA, Culvenor JG, Bottomley SP, Masters CL, Barnham KJ, Hill AF. 2005. Dopamine promotes alpha-synuclein aggregation into SDS-resistant soluble oligomers via a distinct folding pathway. FASEB J 19(8):1377-1379.
Abstract: Dopamine (DA) and alpha-synuclein (alpha-SN) are two key molecules associated with Parkinson's disease (PD). We have identified a novel action of DA in the initial phase of alpha-SN aggregation and demonstrate that DA induces alpha-SN to form soluble, SDS-resistant oligomers. The DA: alpha-SN oligomeric species are not amyloidogenic as they do not react with thioflavin T and lack the typical amyloid fibril structures as visualized with electron microscopy. Circular dichroism studies indicate that in the presence of lipid membranes DA interacts with alpha-SN, causing an alteration to the structure of the protein. Furthermore, DA inhibited the formation of iron-induced alpha-SN amyloidogenic aggregates, suggesting that DA acts as a dominant modulator of alpha-SN aggregation. These observations support the paradigm emerging for other neurodegenerative diseases that the toxic species is represented by a soluble oligomer and not the insoluble fibril.

Callio J, Oury TD, Chu CT. 2005. Manganese superoxide dismutase protects against 6-hydroxydopamine injury in mouse brains. J Biol Chem 280(18): 18536-18542.
Abstract: Dopaminergic neurons of the substantia nigra are susceptible to toxin-based insults. Intrastriatal injection of 6-hydroxydopamine results in selective toxicity to these neurons. A mechanistic role for reactive oxygen species is supported by observations that antioxidants confer protection from 6-hydroxydopamine. Although cell culture studies have suggested extracellular or nonmitochondrial mechanisms in 6-hydroxydopamine

toxicity, the compartmentalization of oxidative injury mechanisms is incompletely defined in vivo. Transgenic mice overexpressing mitochondrial manganese superoxide dismutase or extracellular superoxide dismutase received unilateral intrastriatal injections of 6-hydroxydopamine. Mice that overexpress manganese superoxide dismutase showed significantly smaller striatal lesions than littermate controls. There were no differences in nonspecific striatal injury associated with contralateral vehicle injection. Manganese superoxide dismutase overexpression also protected against loss of neuronal cell bodies in the substantia nigra. In contrast, mice overexpressing extracellular superoxide dismutase showed no protection from 6-hydroxydopamine toxicity in either brain region. Protection of the nigrostriatal system by overexpression of manganese superoxide dismutase supports a role for mitochondrially derived superoxide in 6-hydroxydopamine toxicity. Mitochondrial oxidative stress appears to be a common mechanism among diverse models of Parkinson disease, whether involving toxins, mutated genes, or cybrid cells containing patient mitochondria. Antioxidant therapies that target this subcellular compartment may prove promising.

Calabrese V, Lodi R, Tonon C, D'agata V, Sapienza M, Scapagnini G, Mangiameli A, Pennisi G, Stella AMG, Butterfield DA. 2005. Oxidative stress, mitochondrial dysfunction and cellular stress response in Friedreich's ataxia. J Neurol Sci 233(1-2):145-162.
Abstract: There is significant evidence that the pathogenesis of several neurodegenerative diseases, including Parkinson's disease, Alzheimer's disease, Friedreich's ataxia (FRDA), multiple sclerosis and amyotrophic lateral sclerosis, may involve the generation of reactive oxygen species (ROS) and/or reactive nitrogen species (RNS) associated with mitochondrial dysfunction. The mitochondrial genome may play an essential role in the pathogenesis of these diseases, and evidence for mitochondria being a site of damage in neurodegenerative disorders is based in part on observed decreases in the respiratory chain complex activities in Parkinson's, Alzheimer's, and Huntington's disease. Such defects in respiratory complex activities, possibly associated with oxidant/antioxidant imbalance, are thought to underlie defects in energy metabolism and induce cellular degeneration. The precise sequence of events in FRDA pathogenesis is uncertain. The impaired intramitochondrial metabolism with increased free iron levels and a defective mitochondrial respiratory chain, associated with increased free radical generation and oxidative damage, may be considered possible mechanisms that compromise cell viability. Recent evidence suggests that frataxin might detoxify ROS via activation of glutathione peroxidase and elevation of thiols, and in addition, that decreased expression of frataxin protein is associated with FRDA. Many approaches have been undertaken to understand FRDA, but the heterogeneity of the etiologic factors makes it difficult to define the clinically most important factor determining the onset and progression of the disease. However, increasing evidence indicates that factors such as oxidative stress and disturbed protein metabolism and their interaction in a vicious cycle are central to FRDA pathogenesis. Brains of FRDA patients undergo many changes, such as disruption of protein synthesis and degradation, classically associated with the heat shock response, which is one form of stress response. Heat shock proteins are proteins serving as molecular chaperones involved in the protection of cells from various forms of stress. In the central nervous system, beat shock protein (HSP) synthesis is induced not only after hyperthermia, but also following alterations in the intracellular redox environment. The major neurodegenerative diseases, Alzheimer's disease (AD), Parkinson's disease (PD), amyotrophic lateral sclerosis (ALS), multiple sclerosis (MS), Huntington's disease (HD) and FRDA are all associated with the presence of abnormal proteins. Among the various HSPs, HSP32, also known as heme oxygenase I (HO-1), has received considerable attention, as it has been recently demonstrated that HO-I induction, by generating the vasoactive molecule carbon monoxide and the potent antioxidant bilirubin, could represent a protective system potentially active against brain oxidative injury. Given the broad cytoprotective properties of the heat shock

response there is now strong interest in discovering and developing pharmacological agents capable of inducing the heat shock response. This may open up new perspectives in medicine, as molecules inducing this defense mechanism appear to be possible candidates for novel cytoprotective strategies. In particular, manipulation of endogenous cellular defense mechanisms, such as the heat shock response, through nutritional antioxidants, pharmacological compounds or gene transduction, may represent an innovative approach to therapeutic intervention in diseases causing tissue damage, such as neurodegeneration. (c) 2005 Elsevier B.V. All rights reserved.

Caban-Holt A, Mattingly M, Cooper G, Schmitt FA. 2005. Neurodegenerative memory disorders: A potential role of environmental toxins. Neurol Clin 23 (2):485-+.
Abstract: This article summarizes research literature that evaluates putative environmental exposure to toxins and their potential association with neurodegenerative disorders. Included are the environmental toxins such as solvents, metals, pesticides, magnetic field exposure, and smoking. Because most information about toxins and neurodegeneration is derived from laboratory models and epidemiologic research, causality is difficult to infer when these data are generalized to populations or applied to individual patients.

Brans R, Dickel H, Bruckner T, Coenraads PJ, Heesen M, Merka HF, Blomeke B. 2005. MnSOD polymorphisms in sensitized patients with delayed-type hypersensitivity reactions to the chemical allergen para-phenylene diamine: A case-control study. Toxicology 212(2-3):148-154.
Abstract: Dyes such as para-phenylene diamine (PPD) or related para-compounds are very common contact sensitizers in man. The corresponding contact dermatitis in sensitized individuals is a complex and common illness associated with considerable morbidity and social cost. It has been found that oxidative stress from reactive oxygen species (ROS) may play an important role in the pre-immunological phase of allergic contact dermatitis to PPD. Manganese superoxide dismutase (MnSOD) is one of the primary enzymes that directly scavenge potential harmful oxidizing species. A valine (Val) to alanine (Ala) substitution at amino acid -9, occurring in the MnSOD gene, has been associated with various disease risk. The aim of our study was to investigate possible associations of the MnSOD 47 T > C genotype in exon 2 (Ala-9Val) and the 339 T > C genotype in exon 3 (Ile58Thr) with contact sensitization to PPD in humans in a case-control study. The study was performed in 157 unrelated cases and 201 age- and gender-matched controls. The MnSOD genotypes were determined using LightCycler allele discrimination assays. No heterozygous (CT) or homozygous carriers (TT) for the Ile58Thr polymorphism were found. The frequency for the C allele of the Ala-9Val polymorphism was 51% (79/157) in cases and 49% (107/201) in controls. Homozygous CC carriers (Ala/Ala) were 27% (43/157) in cases and 23% (46/201) in controls (odds ratio [OR], 1.3; 95% confidence interval [CI], 0.8-2.1). Stratification into subgroups based on gender and age limited the association to females. Increased risk among homozygous CC carriers (Ala/Ala) was only found in the group of older females (over 45 years, 25% versus 18%; OR, 1.5; 95% CI, 0.7-2.34). These data suggest that the C (Ala) allele of MnSOD modifies contact dermatitis risk among older females, but is not an independent susceptibility factor for contact sensitization to PPD. (c) 2005 Elsevier Ireland Ltd. All rights reserved.

Bernhard D, Rossmann A, Wick G. 2005. Metals in cigarette smoke. Iubmb Life 57(12):805-809.
Abstract: Metals are vital for a huge number of physiological processes in the human body, but can also destroy health when the concentration is not within the physiologically favourable range. Cigarette smoking interferes with the carefully controlled metal homeostasis of the human body. This review focuses on the consequences of metal delivery to the human body by cigarette smoking and discusses the body's responses. The metal content of tobacco plants, smoke, the circulation, and various organs is

discussed. Finally, we link individual cigarette smoke contained metals to the genesis of human diseases.

Barriere G, Cazalets JR, Bioulac B, Tison F, Ghorayeb I. 2005. The restless legs syndrome. Prog Neurobiol 77(3):139-165.
Abstract: The restless legs syndrome (RLS) is one of the commonest neurological sensorimotor disorders at least in the Western countries and is often associated with periodic limb movements (PLM) during sleep leading to severe insomnia. However, it remains largely underdiagnosed and its underlying pathogenesis is presently unknown. Women are more affected than men and early-onset disease is associated with familial cases. A genetic origin has been suggested but the mode of inheritance is unknown. Secondary causes of RLS may share a common underlying pathophysiology implicating iron deficiency or misuse. The excellent response to dopaminegic drugs points to a central role of dopamine in the pathophysiology of RLS. Iron may also represent a primary factor in the development of RLS, as suggested by recent pathological and brain imaging studies. However, the way dopamine andiron, and probably other compounds, interact to generate the circadian pattern in the occurrence of RLS and PLM symptoms remains unknown. The same is also the case for the level of interaction of the two compounds within the central nervous system (CNS). Recent electrophysiological and animals studies suggest that complex spinal mechanisms are involved in the generation of RLS and PLM symptomatology. Dopamine modulation of spinal reflexes through dopamine D3 receptors was recently highlighted in animal models. The present review suggests that RLS is a complex disorder that may result from a complex dysfunction of interacting neuronal networks at one or several levels of the CNS and involving numerous neurotransmitter systems. (c) 2005 Elsevier Ltd. All rights reserved.

Barreto WJ, Barreto SRG, Ponzoni S, Kawano Y, Di Mauro E, Magosso HA, Silva WP. 2005. Preparation and characterization of a stable semiquinone-iron complex. Monatshefte Fur Chemie 136(5):701-712.
Abstract: Dopamine oxidation by iron oxide (Fe2O3) was studied in the presence and absence of sodium thiosulfate in aqueous medium around pH 7 by UV-Vis spectroscopy. The pH changes from 6 to 8 indicate that the dopamine oxidation process has occurred producing an anionic semiquinone radical which appears after ca. 100 hours presenting bands at 309 and 337 nm. It forms a stable compound with Fe(III) released by the iron oxide. The complex [CTA][Fe(SQ)(2)(CAT)], where SQ=semiquinone, CAT=catecholate, and CTA =cetyltrimethyl ammonium cation, was isolated by precipitation with cetyltrimethylammonium bromide and was characterized through EPR, Raman and IR spectroscopies. The EPR spectrum presented two intense bands, one with g = 2.003 assigned to o-semiquinone and the other with g = 4.274 characteristic for high spin Fe (III) approaching an octahedral symmetry. The most intense Raman resonance band occurs at 1360 cm(-1) assigned to nu(C-1-C-2) and at 1575 cm(-1) to nu(C-C)ring of the o-semiquinone. The 0(2) dissolved in solution is mainly responsible for the dopamine oxidation when sodium thiosulfate is present. A thermal decomposition mechanism based on the thermogravimetric curves (TG) was proposed. These results suggest that iron can participate in the degenerative process of the dopaminergic nigral neurons. Its role seems to be its coordination with the dopamine oxidation products as o-semiquinone and catecholate which could damage neurons giving rise to parkinsonism.

Babincova M, Babinec P. 2005. Dopamine mediated iron release from ferritin is enhanced at higher temperatures: Possible implications for fever induced Parkinson's disease. Journal of Magnetism and Magnetic Materials 293(1. Sp. Iss. Si):341-344.
Abstract: A new molecular mechanism is proposed to explain the pathogenesis of fever-induced Parkinson's disease. This proposal is based on dopamine and 6-hydroxydopamine-mediated free iron release from ferritin magnetic nanoparticles, which is enhanced at higher temperatures, and which may lead to substantial peroxidation and injury of lipid

biomembranes of the substantia nigra in the brain. (c) 2005 Elsevier B.V. All rights reserved.

Aschner M, Erikson KM, Dorman DC. 2005. Manganese dosimetry: Species differences and implications for neurotoxicity. Crit Rev Toxicol 35(1):1-32.
Abstract: Manganese (Mn) is an essential mineral that is found at low levels in food, water, and the air. Under certain high-dose exposure conditions, elevations in tissue manganese levels can occur. Excessive manganese accumulation can result in adverse neurological, reproductive, and respiratory effects in both laboratory animals and humans. In humans, manganese-induced neurotoxicity (manganism) is the overriding concern since affected individuals develop a motor dysfunction syndrome that is recognized as a form of parkinsonism. This review primarily focuses on the essentiality and toxicity of manganese and considers contemporary studies evaluating manganese dosimetry and its transport across the blood-brain barrier, and its distribution within the central nervous system (CNS). These studies have dramatically improved our understanding of the health risks posed by manganese by determining exposure conditions that lead to increased concentrations of this metal within the CNS and other target organs. Most individuals are exposed to manganese by the oral and inhalation routes of exposure; however, parenteral injection and other routes of exposure are important. Interactions between manganese and iron and other divalent elements occur and impact the toxicokinetics of manganese, especially following oral exposure. The oxidation state and solubility of manganese also influence the absorption, distribution, metabolism, and elimination of manganese. Manganese disposition is influenced by the route of exposure. Rodent inhalation studies have shown that manganese deposited within the nose can undergo direct transport to the brain along the olfactory nerve. Species differences in manganese toxicokinetics and response are recognized with nonhuman primates replicating CNS effects observed in humans while rodents do not. Potentially susceptible populations, such as fetuses, neonates, individuals with compromised hepatic function, individuals with suboptimal manganese or iron intake, and those with other medical states (e.g., pre-parkinsonian state, aging), may have altered manganese metabolism and could be at greater risk for manganese toxicity.

[Anon]. 2005. Increased expression of DMT1 may be the cause of elevated iron in SN of Parkinsonian animal models. Cell Biol Int 29(10):S12.

Andre C, Truong TT, Robert JF, Guillaume YC. 2005. Effect of metals on herbicides-alpha-synuclein association: A possible factor in neurodegenerative disease studied by capillary electrophoresis. Electrophoresis 26(17):3256-3264.
Abstract: The aggregation of a-synuclein in the dopaminergic neurons of the substantia nigra is a critical step in the Parkinson's disease (PD). The etiology of the disease is unknown but recent epidemiological and experimental studies have renewed interest in the hypothesis that environmental factors, especially herbicides and metals, have a role on the pathogenesis of PD. For the first time, the association constants of a-synuclein with five herbicides have been calculated using a capillary electrophoresis (CE) method. In addition, the effect of a number of metals on this binding has been investigated. It appears that the herbicides preferentially bind to a partially folded intermediate conformation of a-synuclein induced by manganese, aluminium, cadmium, copper and zinc. Then, metal increases the synuclein-herbicide association. However, this study shows contrasting actions with the antibiotic rifampicin and magnesium addition leading to a decrease of the alpha-synuclein-herbicide interaction even if other metals are present in the bulk solvent. Considering epidemiological studies, all these results suggest an underlying molecular basis for PD and related body diseases.

Andoh T, Chock PB, Murphy DL, Chiueh CC. 2005. Role of the redox protein thioredoxin in cytoprotective mechanism evoked by (-)-deprenyl. Mol Pharmacol 68(5):1408-1414.

Abstract: Through the inhibition of monoamine oxidase type B (MAO-B), (-)-deprenyl (selegiline) prevents the conversion of 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) to the toxic metabolite 1-methyl-4- phenylpyridinium ion (MPP+) and also prevents the neurotoxicity in the dopaminergic neurons in animal models. Cumulative observations suggest that selegiline may also protect against MPP+-induced neurotoxicity, possibly through the induction of pro-survival genes. We have observed that thioredoxin (Trx) mediates the induction of mitochondrial manganese superoxide dismutase (MnSOD) and Bcl-2 during preconditioning-induced hormesis. We therefore investigated whether the redox protein Trx plays any role in the neuroprotective mechanism of selegiline against MPP+-induced cytotoxicity in human SH-SY5Y neuroblastoma cells and also in primary neuronal cultures of mouse midbrain dopaminergic neurons. After confirming that selegiline protects against MPP+-induced cytotoxicity, we observed further that selegiline, at 1 mu M or less, induced Trx for protection against oxidative injury caused by MPP+. The induction of Trx was blocked by protein kinase A (PKA) inhibitor and mediated by a PKA-sensitive phospho-activation of mitogen-activated protein (MAP) kinase Erk1/2 and the transcription factor c-Myc. Selegiline-induced Trx and associated neuroprotection were concomitantly blocked by the antisense against Trx mRNA, but not the sense or antisense mutant phosphothionate oligonucleotides, not only in human SH-SY5Y cells but also in mouse primary neuronal culture of midbrain dopaminergic neurons. Furthermore, the redox cycling of Trx may mediate the protective action of selegiline because the inhibition of Trx reductase by 1-chloro-2,4-dinitrobenzene ameliorated the effect of selegiline. Trx (1 mu M) consistently increased the expression of mitochondrial proteins MnSOD and Bcl-2, supporting cell survival (Andoh et al., 2002). In conclusion, without modifying MAO-B activity, selegiline augments the gene induction of Trx, leading to elevated expression of antioxidative MnSOD and antiapoptotic Bcl-2 proteins for protecting against MPP+-induced neurotoxicity.

Alvarez E, Zhou WB, Witta SE, Freed CR. 2005. Characterization of the Bex gene family in humans, mice, and rats. Gene 357(1):18-28.
Abstract: To better understand the development of ventral mesencephalic dopamine neurons, we performed subtractive hybridization screens to find ventral mesencephalic genes expressed at rat embryonic day 10 when these neurons begin to differentiate. The most commonly identified genes in these screens were members of the Bex (Brain expressed X-linked) gene family, rat Bex1 (Rex3), and a novel gene, rat Bex4. After identifying these genes, we then sought to characterize the Bex gene family. Two additional novel Bex genes (human Bex5 and mouse Bex6) were discovered through genomic databases. Bex5 is present in humans and monkeys, but not rodents, while Bex6 exists in mice, but not humans. Bex4 and Bex5 are localized to the X chromosome, are expressed in brain, and are similar in sequence. Bex4 and Bex5 are 54% and 56% identical to human Bex3 (pHGR74, NADE). Mouse Bex6 is on chromosome 16 and is 67% identical to mouse Bex4. Human Bex gene expression was studied with tissue expression arrays probed with specific oligonucleotides. Human Bex1 and Bex2 have similar expression patterns in the central nervous system with high levels in pituitary, cerebellum, and temporal lobe, and Bex1 is widely expressed outside of the central nervous system with high expression in the liver. Human Bex4 is highly expressed in heart, skeletal muscle, and liver, while Bex3 and Bex5 are more widely expressed. The subcellular localization of the Bex proteins varies from nuclear (rat Bex1) to cytoplasmic (rat Bex3, human Bex5, and mouse Bex6) and to both nuclear and cytoplasmic (rat Bex2 and rat Bex4). Rat Bex3, rat Bex4, human Bex5, and mouse Bex6 are degraded by the proteasome, while rat Bex1 or Bex2 are not. Rat Bex3 protein can likely bind transition metals through a histidine-rich domain. Because this gene family was originally named Bex and because these genes are unified by sequence similarity and gene structure, we believe the Bex nomenclature should prevail over nomenclature based on function (NADE) that has not been extended to the other Bex genes. We conclude that the Hex gene family members are highly homologous but differ in their expression patterns, subcellular

localization, and degradation by the proteasome. (c) 2005 Elsevier B.V. All rights reserved.

Akyol O, Yanik M, Elyas H, Namli M, Canatan H, Akin H, Yuce H, Yilmaz HR, Tutkun H, Sogut S, Herken H, Ozyurt H, Savas HA, Zoroglu SS. 2005. Association between Ala-9Val polymorphism of Mn-SOD gene and schizophrenia. Progress in Neuro-Psychopharmacology & Biological Psychiatry 29(1):123-131.
Abstract: Reactive oxygen species (ROS) have been suggested to play an important role in physiopathology of schizophrenia. The major intracellular antioxidant enzymes, copper-zinc superoxide dismutase in the cytoplasm and manganese superoxide dismutase (Mn-SOD) in the mitochondria, rapidly and specifically reduce superoxide radicals to hydrogen peroxide. Polymorphisms in the genes encoding antioxidant enzymes should therefore result in predisposition to schizophrenia. The present study was performed to assess; whether there is a genetic association between a functional polymorphism (Ala-9Val) in the human Mn-SOD gene in schizophrenic patients (n=153) and healthy controls (n=196) using a PCR/RFLP method. Significant differences in the genotypic distribution between schizophrenics and controls were observed. Genotypic distribution with 14 (9.2%) Ala/Ala, 106 (69.3%) Ala/Val and 33 (21.6%) Val/Val subjects, in schizophrenia was different from those of controls with 46 (23.5%), 83 (42.3%) and 67 (34.21/6), respectively ($p<0.0001$). When the patents with schizophrenia were divided into the subgroups as disorganized, paranoid and residual. there was a significant difference In genotypic distribution among the subgroups ($chi(2)=11.35$, $df=4$ $p=0.023$). This association between -9Ala Mn-SOD allele and schizophirenia suggests that -9Ala variant may have a contribution in the physiopathogenesis of schizophrenia. Further investigations are warranted in larger populations with other susceptible genes that might be associated with schizophrenia. (C) 2004 Published by Elsevier Inc.

Ajjimaporn A, Swinscoe J, Shavali S, Govitrapong P, Ebadi M. 2005. Metallothionein provides zinc-mediated protective effects against methamphetamine toxicity in SK-N-SH cells. Brain Res Bull 67(6):466-475.
Abstract: Methamphetamine (METH) is a drug of abuse and neurotoxin that induces Parkinson's-like pathology after chronic usage by targeting dopaminergic neurons. Elucidation of the intracellular mechanisms that underlie METH-induced dopaminergic neuron toxicity may help in understanding the mechanism by which neurons die in Parkinson's disease. In the present study, we examined the role of reactive oxygen species (ROS) in the METH-induced death of human dopaminergic SK-N-SH cells and further assessed the neuroprotective effects of zinc and metallothionein (MT) against METH-induced toxicity in culture. METH significantly increased the production of reactive oxygen species, decreased intracellular ATP levels and reduced the cell viability. Pre-treatment with zinc markedly prevented the loss of cell viability caused by METH treatment. Zinc Pre-treatment mainly increased the expression of metallothionein and prevented the generation of reactive oxygen species and ATP depletion caused by METH. Chelation of zinc by CaEDTA caused a significant decrease in MT expression and loss of protective effects of MT against METH toxicity. These results suggest that zinc-induced MT expression protects dopaminergic neurons via preventing the accumulation of toxic reactive oxygen species and halting the decrease in ATP levels. Furthermore, MT may prevent the loss of mitochondrial functions caused by neurotoxins. In conclusion, our study suggests that MT, a potent scavenger of free radicals is neuroprotective against dopaminergic toxicity in conditions such as drug of abuse and in Parkinson's disease. (c) 2005 Published by Elsevier Inc.

Youdim MB, Fridkin M, Zheng H. 2004 Oct. Novel bifunctional drugs targeting monoamine oxidase inhibition and iron chelation as an approach to neuroprotection in Parkinson's disease and other neurodegenerative diseases. J Neural Transm 111(10-11):1455-71.
Abstract: Iron has been shown to accumulates at site where neurons

degenerate in neurodegenerative diseases of Parkinson's disease, Alzheimer's disease, Huntington disease, amyotrophic lateral sclerosis and Friedreich ataxia. Iron is thought to participate or initiate oxidative stress via generation of reactive oxygen species (ROS), such as hydroxyl radical. Iron chelators are neuroprotective and prevent 6-hydroxydoapmine and MPTP dopaminergic neurotoxicity in rats and mice. However, their action on monoamine oxidase (MAO) A and B have not been determined previously since MAO-B inhibitors have been shown to be neuroprotective in cellular and animal models of Parkinson's disease. The chelators 8-hydroxyquinoline, O-phenanthroline, 2,2'-dipyridyl, U74500A and U74600F showed a preference for inhibition of rat brain mitochondrial MAO-A over MAO-B. Their IC(50) ranged from 10(-3) M to 10(-6) M, with 21-amino steroids (U74500A and U74006F) showing a greater selectivity and potency for MAO-A. Desferrioxamine (desferal), a prototype potent iron chelator, exhibited relatively poor MAO inhibitory. The inhibitions of MAO-A and B by 21-amino steroids (Lazaroids) were time dependent and irreversible. Those initiated by 8-hydroxyquinoline, 2,2'-dipyridyl and O-phenanthroline were fully reversible by enzyme dilution experiments. Both Fe(2+) and Fe(3+) reverse the MAO-A and B inhibition induced by the latter chelators, but not those initiated by 21-amino steroids. The data infer that either the inhibition of MAO by 21-amino steroids is either the resultant of their conversion to an irreversible covalently bound ligand or that the iron chelation moiety and MAO inhibitory activity in these compounds are not mutually shared. The results suggest that bifunctional brain penetrable drugs with iron chelating property and MAO inhibitory activity in could be the most feasible approach for neuroprotection in neurodegenerative diseases. Such drug would prevent participation of elevated iron in oxidative stress and formation of reactive hydroxyl radical, via its interaction with H(2)O2 (Fenton chemistry), generated as a consequence MAO and other oxidative enzyme reactions to generative cytotoxic reactive hydroxyl radical. We have now developed several of these compounds with neuroprotective, MAO inhibitory and iron chelating properties from our prototype iron chelators, VK-28 possessing propargylamine moiety of our anti-parkinson drug, rasagiline.

Naito Y, Kuzuhara S. 2004 Sep. [Essential points to differentiate various diseases causing parkinsonism]. Nippon Rinsho 62(9):1608-16.
Abstract: This review article deals with the cardinal features to differentiate various conditions which present with parkinsonism other than Parkinson's disease. Special attention is paid to the distinctive clinical features, laboratory data and neuroimaging findings of frequent diseases as well as important ones including multiple system atrophy, progressive supranuclear palsy, corticobasal degeneration, Lewy body disease, drug-induced parkinsonism, vascular pseudo-parkinsonism, normal pressure hydrocephalus and manganese intoxication due to parenteral nutrition. MRI is useful to diagnose MSA, vascular pseudo-parkinsonism and manganese intoxication. Benzamide derivatives including sulpiride and metoclopramide are the main causes of drug-induced parkinsonism in recent years in Japan.

Schipper HM. 2004 Jul. Brain iron deposition and the free radical-mitochondrial theory of ageing. Ageing Res Rev 3(3):265-301.
Abstract: The central hypothesis of this paper states that oxidative stress, augmented iron deposition, and mitochondrial insufficiency in the ageing and degenerating CNS constitute a single neuropathological 'lesion', and that the advent of one component of this triad obligates the appearance of the others. Evidence in support of this unifying perspective is adduced from human neuropathological studies, experimental paradigms of ageing-associated neurological disorders, and a comprehensive model of astroglial senescence. A pivotal role for the enzyme, heme oxygenase-1 (HO-1) in consolidating this tripartite lesion in the ageing and diseased CNS is emphasized. The data are discussed in the context of a revised 'free radical-mitochondrial-metal' theory of brain ageing, and some scientific and clinical implications of the latter are considered.

Sadrzadeh SM, Saffari Y. 2004 Jun. Iron and brain disorders. Am J Clin Pathol 121 Suppl:S64-70.
Abstract: Iron is the most important element in the body, essential for almost all types of cells, including brain cells. The role of iron in the brain has been known for years. Iron deficiency and iron excess have been associated with pathophysiology of different brain disorders. Iron deficiency has been reported to have a role in brain development and the pathophysiology of restless legs syndrome. Iron accumulation has been related to some neurologic disorders such as Alzheimer disease, Parkinson disease, type I neurodegeneration with brain iron accumulation, and other disorders. Despite years of investigation, the reason for iron imbalance in the brain is not known. It also is not known whether the accumulation of iron in the brain is primary or secondary to development of neurodegenerative disorders. This review summarizes the present knowledge on the role of iron in human brain disorders.

Youdim MB, Stephenson G, Ben Shachar D. 2004 Mar. Ironing iron out in Parkinson's disease and other neurodegenerative diseases with iron chelators: a lesson from 6-hydroxydopamine and iron chelators, desferal and VK-28. Ann N Y Acad Sci 1012:306-25 .
Abstract: In Parkinson's disease (PD) and its neurotoxin-induced models, 6-hydroxydopamine (6-OHDA) and N-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP), significant accumulation of iron occurs in the substantia nigra pars compacta. The iron is thought to be in a labile pool, unbound to ferritin, and is thought to have a pivotal role to induce oxidative stress-dependent neurodegeneration of dopamine neurons via Fenton chemistry. The consequence of this is its interaction with H(2)O(2) to generate the most reactive radical oxygen species, the hydroxyl radical. This scenario is supported by studies in both human and neurotoxin-induced parkinsonism showing that disposition of H(2)O(2) is compromised via depletion of glutathione (GSH), the rate-limiting cofactor of glutathione peroxide, the major enzyme source to dispose H(2)O(2) as water in the brain. Further, radical scavengers have been shown to prevent the neurotoxic action of the above neurotoxins and depletion of GSH. However, our group was the first to demonstrate that the prototype iron chelator, desferal, is a potent neuroprotective agent in the 6-OHDA model. We have extended these studies and examined the neuroprotective effect of intracerebraventricular (ICV) pretreatment with the prototype iron chelator, desferal (1.3, 13, 134 mg), on ICV induced 6-OHDA (250 micro g) lesion of striatal dopamine neurons. Desferal alone at the doses studied did not affect striatal tyrosine hydroxylase (TH) activity or dopamine (DA) metabolism. All three pretreatment (30 min) doses of desferal prevented the fall in striatal and frontal cortex DA, dihydroxyphenylacetic acid, and homovalinic acid, as well as the left and right striatum TH activity and DA turnover resulting from 6-OHDA lesion of dopaminergic neurons. A concentration bell-shaped neuroprotective effect of desferal was observed in the striatum, with 13 micro g being the most effective. Neither desferal nor 6-OHDA affected striatal serotonin, 5-hydroxyindole acetic acid, or noradrenaline. Desferal also protected against 6-OHDA-induced deficit in locomotor activity, rearing, and exploratory behavior (sniffing) in a novel environment. Since the lowest neuroprotective dose (1.3 micro g) of desferal was 200 times less than 6-OHDA, its neuroprotective activity may not be attributed to interference with the neurotoxin activity, but rather iron chelation. These studies led us to develop novel brain-permeable iron chelators, the VK-28 series, with iron chelating and neuroprotective activity similar to desferal for ironing iron out from PD and other neurodegenerative diseases, such as Alzheimer's disease, Friedreich's ataxia, and Huntington's disease.

Jimenez Del Rio M, Moreno S, Garcia-Ospina G, Buritica O, Uribe CS, Lopera F, Velez-Pardo C. 2004 Mar. Autosomal recessive juvenile parkinsonism Cys212Tyr mutation in parkin renders lymphocytes susceptible to dopamine- and iron-mediated apoptosis. Mov Disord 19(3):324-30.
Abstract: Mutations in parkin are implicated in the pathogenesis of autosomal recessive juvenile parkinsonism (AR-JP) disease. We show that

homozygote Cys212Tyr parkin mutation in AR-JP patients renders lymphocytes sensitive to dopamine, iron and hydrogen peroxide stimuli. Indeed, dopamine-induced apoptosis by four alternative mechanisms converging on caspase-3 activation and apoptotic morphology: (1) NF-kappaB-dependent pathway; mitochondrial dysfunction either by (2) H(2)O (2) or (3) hydroxyl exposure and (4) increase of unfolded-protein stress. We also demonstrate that 17beta-estradiol and testosterone prevent homozygote lymphocytes from oxidative stressors-evoked apoptosis. These results may contribute to understanding the relationship between genetic and environmental factors and iron in AR-JP.

Yoshida T, Suzuki G, Nibuya M, Sano SY, Nomura S. 2004 Feb. Parkinsonism induced by atypical neuroleptics in a patient with severe iron deficiency. Nihon Shinkei Seishin Yakurigaku Zasshi 24(1):29-31.
Abstract: Although still controversial, iron deficiency has been indicated as one of the risk factors for developing neuroleptic-induced extrapyramidal symptoms (EPSs), including akathisia, dystonia, and neuroleptic malignant syndrome. Here we report our experience of iron supplementation and alternating neuroleptics for treating Parkinsonism in a schizophrenic female patient having severe iron deficient anemia.

Shachar DB, Kahana N, Kampel V, Warshawsky A, Youdim MB. 2004 Feb. Neuroprotection by a novel brain permeable iron chelator, VK-28, against 6-hydroxydopamine lession in rats. Neuropharmacology 46(2):254-63.
Abstract: Significant increase in iron occurs in the substantia nigra pars compacta of Parkinsonian subjects, and in 6-hydroxydopamine (6-OHDA) treated rats and monkeys. This increase in iron has been attributed to its release from ferritin and is associated with the generation of reactive oxygen species and the onset of oxidative stress-induced neurodegeneration. Several iron chelators with hydroxyquinoline backbone were synthesized and their ability to inhibit basal as well as iron-induced mitochondrial lipid peroxidation was examined. The neuroprotective potential of the brain permeable iron chelator, VK-28 (5-[4-(2-hydroxyethyl) piperazine-1-ylmethyl]-quinoline-8-ol), injected either intraventricularly (ICV) or intraperitoneally (IP), to 6-OHDA lesioned rats was investigated. VK-28 inhibited both basal and Fe/ascorbate induced mitochondrial membrane lipid peroxidation, with an IC(50) (12.7 microM) value comparable to that of the prototype iron chelator, desferal, which does not cross the blood brain barrier. At an ICV pretreatment dose as low as 1 microg, VK-28 was able to completely protect against ICV 6-OHDA (250 microg) induced striatal dopaminergic lesion, as measured by dopamine (DA), dihydroxyphenylacetic acid (DOPAC) and homovanilic acid (HVA) levels. IP injection of rats with VK-28 (1 and 5 mg/kg) daily for 10 and 7 days, respectively, demonstrated significant neuroprotection against ICV 6-OHDA at the higher dose, with 68% protection against loss of dopamine at 5mg/kg dosage of VK-28. The present study is the first to show neuroprotection with a brain permeable iron chelator. The latter can have implications for the treatment of Parkinson's disease and other neurodegenerative diseases (Alzheimer's disease, Friedreich ataxia, aceruloplasminemia, Hallervorden Spatz syndrome) where abnormal iron accumulation in the brain is thought to be associated with the degenerative processes.

Beuter A, Edwards R. 2004 Feb. Effect of chronic exposure to methylmercury on eye movements in Cree subjects. Int Arch Occup Environ Health 77(2): 97-107.
Abstract: OBJECTIVES: To examine the effect of chronic exposure to methylmercury on eye movements (pursuit, fixation and dynamic saccades) in Cree subjects from Northern Quebec. METHODS: Eye movements were recorded in a group of Cree subjects (n=36) exposed chronically to methylmercury, a group of patients with Parkinson's disease (PD) (n=21), and a group of control subjects (n=30) by use of an infrared eye-movement recording system. Pursuit, fixation, and prompted and remembered saccades were recorded twice for both eyes in the horizontal and vertical axes. Blinks were removed, and data were calibrated.

RESULTS: Analyses of variance revealed significant differences for all characteristics examined for fixation and pursuit, and for some characteristics in dynamic saccades. These differences arose sometimes from the Cree group, sometimes from the PD group and sometimes from both groups. CONCLUSIONS: The results suggest that eye movements of Cree subjects exposed to methylmercury are qualitatively different from those of both control subjects and patients with PD. Comparisons between more-exposed and less-exposed Cree subjects matched for age with control subjects also showed significant differences for fixation, pursuit and dynamic saccades. The average scores of the more-exposed group were clearly separated from those of the less-exposed and control groups for characteristics of fixation and pursuit, and for accuracy and sharpness of prompted saccades. This trend was less clear in other results where a possible effect of mercury exposure could not be distinguished from a possible cultural effect. Further studies should focus on the most discriminating characteristics for the Cree group, such as measures of accuracy and coherence in all tests and sharpness of saccades.

Yokel RA, Crossgrove JS. 2004 Jan. Manganese toxicokinetics at the blood-brain barrier. Res Rep Health Eff Inst (119):7-58; discussion 59-73.
Abstract: Increased manganese (Mn) use in manufacturing and in gasoline has raised concern about Mn-induced parkinsonism. Previous research indicated carrier-mediated brain entry but did not assess brain efflux. Using in situ rat brain perfusion, we studied influx across the blood-brain barrier (BBB*) of three predominant plasma Mn species available to enter the brain: Mn2+, Mn citrate, and Mn transferrin. Our results suggested transporter-mediated uptake of these species. The uptake rate was greatest for Mn citrate. Our results using the brain efflux index method suggested that diffusion mediates distribution from rat brain to blood. To characterize the carriers mediating brain Mn uptake, we used rat erythrocytes, an immortalized murine BBB cell line (b.End5), primary bovine brain endothelial cells (bBMECs), and Sprague Dawley and Belgrade rats. Studies with bBMECs and b.End5 cells suggested concentrative brain Mn2+ and Mn citrate uptake, respectively, consistent with carrier-mediated uptake. Mn2+ uptake positively correlated with pH, suggesting mediation by an electromotive force. Mn2+ uptake was not inhibited by iron or the absence of divalent metal transporter 1 (DMT-1) expression, suggesting an iron-transporter-independent mechanism. Mn2+ uptake inversely correlated with calcium and was affected by calcium channel modulators, suggesting a role for calcium channels. Rat erythrocyte results suggested monocarboxylate transporter 1 (MCT1) and anion exchange transporters do not mediate Mn citrate brain uptake. Considering carrier-mediated brain influx (but not efflux), repeated excessive Mn exposure should produce brain accumulation. Further work is necessary to identify the specific transporter or transporters mediating Mn distribution across the BBB.

Yasui M. 2004 Jan. [Calcium and the degenerative neurological diseases]. Clin Calcium 14(1):110-7.
Abstract: A condition of unbalanced minerals was found in soil and drinking water from three amyotrophic lateral sclerosis (ALS) foci on Guam, in the Kii Peninsula and in West New Guinea with a low concentration of calcium and magnesium coupled with a high concentration of aluminum and manganese. The current epidemiological studies in the Western Pacific including the Kii Peninsula of Japan, have suggested that environmental factors contribute to the pathogenetic process of ALS and parkinsonism-dementia (PD). Six Kii cases with ALS showed higher Ca and lower Mg contents in the central nervous system (CNS) tissues than those of neurologically normal controls. We subsequently designed an animal study to experimentally ascertain the mineral or metal deposition in CNS tissues under various dietary regimens using rats. The experimental results suggest that unbalanced minerals and/or metals lead to the accumulation not only of Ca, but also Mn, and Al, and diminution of Mg and Zn in CNS tissues of rats and humans on these dietary regimens, with implication for long-term neuronal degeneration and accumulating CNS deficit.

Zucconi M, Ferini-Strambi L. 2004. Epidemiology and clinical findings of restless legs syndrome. Sleep Medicine 5(3):293-299.
Abstract: Restless legs syndrome (RLS) is a sensory-motor disorder characterized by discomfort of and urge to move the legs, primarily during rest or inactivity, partial or total relief with movement, with presence or worsening exclusively in the evening. It is a relatively common but frequently unrecognized disorder, with a prevalence ranging from 2.5 to 15% of the general population, increasing with age and with a female preponderance. The diagnosis is clinical but polysomnography is useful to determine its profound impact on sleep (difficulties in sleep onset, maintaining sleep during the night, and sleep fragmentation) and for the evidence of periodic legs movements during sleep and wake. RLS is generally idiopathic. with familial association in 40-60% of the cases, but may also be symptomatic of such associated conditions (secondary forms) as peripheral neuropathies, uremia, iron deficiency (with or without anemia), diabetes, Parkinson's disease and pregnancy. Response to dopaminergic drugs indicates that dopamine receptors are implicated, and although much progress has been made ill diagnosis and treatment in the last decade, more is needed for complete elucidation of the etiology and pathophysiology of RLS. (C) 2004 Elsevier B.V. All rights reserved.

Zucca FA, Giaveri G, Gallorini M, Albertini A, Toscani M, Pezzoli G, Lucius R, Wilms H, Sulzer D, Ito S, Wakamatsu K, Zecca L. 2004. The neuromelanin of human substantia nigra: Physiological and pathogenic aspects. Pigment Cell Res 17(6):610-617.
Abstract: Neuromelanin (NM) accumulates as a function of age in normal human substantia nigra (SN) but is relatively depleted in the SN of patients with Parkinson disease (PD). Several studies have been performed to further our understanding of the role of NM in neuronal aging and neurodegenerative mechanisms of PD. To this purpose, NM from human SN was isolated and its structure and molecular interactions were investigated. Cysteinyl-dopamine was shown to be one precursor of NM synthesis. A striking affinity of NM for specific metals, lipids, drugs and pesticides was found in vitro, and in animal and human brain postmortem studies. Because of these affinities, NM seems to play a protective role in the human brain by blocking toxic molecules. On the other hand, experiments in cell culture indicate that NM can activate microglia, eliciting the release of cytotoxic factors that can induce neurodegeneration.

Zhou Y, Shie FS, Piccardo P, Montine TJ, Zhang J. 2004. Proteasomal inhibition induced by manganese ethylene-bis-dithiocarbamate: Relevance to Parkinson's disease. Neuroscience 128(2):281-291.
Abstract: Maneb, a widely used fungicide, has been associated with Parkinsonism in humans. In experimental models, maneb and its major active element, manganese ethylenebis-dithiocarbamate (Mn-EBDC) cause selective nigrostriatal neurodegeneration in mice and in rats, respectively. To investigate the mechanisms underlying this neurodegeneration, we studied the effects of Mn-EBDC on proteasomal function, which is decreased in patients with Parkinson's disease (PD), in a dopaminergic neuronal cell line (MES 23.5 or MES). The results demonstrated that exposure of MES cells to 6 muM Mn-EBDC for 7 days produced not only significant neurotoxicity but also inhibition of proteasomal chymotrypsin-like and postglutamyl peptidase activities. Proteasomal dysfunction was accompanied by formation of cytoplasmic inclusions that were positive for a-synuclein immunostaining and significantly increased sodium dodecyl sulfate-insoluble alpha-synuclein aggregation seen by Western blot analysis. In addition, there was a significant increase in oxidative stress, evidenced by elevated total protein carbonyl content, in cells treated with Mn-EBDC. Manipulation of intracellular reduced glutathione levels with N-acetyl-L-cysteine or L-buthionine sulfoximine pretreatment to modulate Mn-EBDC-mediated oxidative stress altered Mn-EBDC-mediated neurotoxicity, proteasomal dysfunction, and alpha-synuclein aggregation in these cells. These data suggest that neurotoxicityinduced by Mn-EBDC is at least partially attributable to Mn-EBDC-mediated proteasomal inhibition, and that the proteasome may be an important target by which environmental

exposure modifies the risk for developing PD in vulnerable populations. (C) 2004 IBRO. Published by Elsevier Ltd. All rights reserved.

Zemke D, Majid A. 2004. The potential of minocycline for neuroprotection in human neurologic disease. Clin Neuropharmacol 27(6):293-298.
Abstract: Minocycline is a member of the tetracycline class of molecules with broad-spectrum antibiotic activity. The unique properties of minocycline result in increased tissue distribution when compared with the other tetracyclines. Of particular interest is the ability of minocycline to diffuse into the central nervous system at clinically effective levels. Aside from its antimicrobial properties, minocycline has been found to have beneficial effects on inflammation, microglial activation, matrix metalloproteinases, nitric oxide production, and apoptotic cell death. Concordantly. tninocycline has been found to have neuroprotective effects in animal models of a number of diseases including stroke, multiple sclerosis, and Parkinson disease. The proven safety of tninocycline over decades of use as an antibiotic suggests that it may have potential for development into an effective treatment of multiple neurologic conditions in humans.

Zecca L, Youdim MBH, Riederer P, Connor JR, Crichton RR. 2004. Iron, brain ageing and neurodegenerative disorders. Nature Reviews Neuroscience 5 (11):863-873.
Abstract: There is increasing evidence that iron is involved in the mechanisms that underlie many neurodegenerative diseases. Conditions such as neuroferritinopathy and Friedreich ataxia are associated with mutations in genes that encode proteins that are involved in iron metabolism, and as the brain ages, iron accumulates in regions that are affected by Alzheimer's disease and Parkinson's disease. High concentrations of reactive iron can increase oxidative-stress induced neuronal vulnerability, and iron accumulation might increase the toxicity of environmental or endogenous toxins. By studying the accumulation and cellular distribution of iron during ageing, we should be able to increase our understanding of these neurodegenerative disorders and develop new therapeutic strategies.

Zecca L, Stroppolo A, Gatti A, Tampellini D, Toscani M, Gallorini M, Giaveri G, Arosio P, Santambrogio P, Fariello RG, Karatekin E, Kleinman MH, Turro N, Hornykiewicz O, Zucca FA. 2004. The role of iron and copper molecules in the neuronal vulnerability of locus coeruleus and substantia nigra during aging. Proc Natl Acad Sci U S A 101(26):9843-9848.
Abstract: In this study, a comparative analysis of metal-related neuronal vulnerability was performed in two brainstem nuclei, the locus coeruleus (LC) and substantia nigra (SN), known targets of the etiological noxae in Parkinson's disease and related disorders. LC and SN pars compacta neurons both degenerate in Parkinson's disease and other Parkinsonisms; however, LC neurons are comparatively less affected and with a variable degree of involvement. In this study, iron, copper, and their major molecular forms like ferritins, ceruloplasmin, neuromelanin (NM), manganese-superoxide dismutase (SOD), and copper/zinc-SOD were measured in LC and SIN of normal subjects at different ages. Iron content in LC was much lower than that in SN, and the ratio heavy-chain ferritin/ iron in LC was higher than in the SN. The NM concentration was similar in LC and SIN, but the iron content in NM of LC was much lower than SN. In both regions, heavy- and light-chain ferritins were present only in glia and were not detectable in neurons. These data suggest that in LC neurons, the iron mobilization and toxicity is lower than that in SIN and is efficiently buffered by NM. The bigger damage occurring in SIN could be related to the higher content of iron. Ferritins accomplish the same function of buffering iron in glial cells. Ceruloplasmin levels were similar in LC and SN, but copper was higher in LC. However, the copper content in NM of LC was higher than that of SN, indicating a higher copper mobilization in LC neurons. Manganese-SOD and copper/zinc-SOD had similar age trend in LC and SN. These results may explain at least one of the reasons underlying

lower vulnerability of LC compared to SN in Parkinsonian syndromes.

Youdim MBH, Stephenson G, Ben Shachar D. 2004. Ironing Iron Out in Parkinson's Disease and Other Neurodegenerative Diseases With Iron Chelators - a Lesson From 6-Hydroxydopamine and Iron Chelators, Desferal and Vk-28Volume 1012. p 306-325. Redox-Active Metals in Neurological Disorders: Annals of the New York Academy of Sciences.
Abstract: In Parkinson's disease (PD) and its neurotoxin-induced models, 6-hydroxydopamine (6-OHDA) and N-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP), significant accumulation of iron occurs in the substantia nigra pars compacta. The iron is thought to be in a labile pool, unbound to ferritin, and is thought to have a pivotal role to induce oxidative stress-dependent neuro-degeneration of dopamine neurons via Fenton chemistry. The consequence of this is its interaction with H2O2 to generate the most reactive radical oxygen species, the hydroxyl radical. This scenario is supported by studies in both human and neurotoxin-induced parkinsonism showing that disposition of H2O2 is compromised via depletion of glutathione (GSH), the rate-limiting cofactor of glutathione peroxide, the major enzyme source to dispose H2O2 as water in the brain. Further, radical scavengers have been shown to prevent the neurotoxic action of the above neurotoxins and depletion of GSH. However, our group was the first to demonstrate that the prototype it-on chelator, desferal, is a potent neuroprotective agent in the 6-OHDA model. We have extended these studies and examined the neuroprotective effect of intracerebraventricular (ICV) pretreatment with the prototype iron chelator, desferal (1.3, 13, 134 mug), on ICV induced 6-OHDA (250 mug) lesion of striatal dopamine neurons. Desferal alone at the doses studied did not affect striatal tyrosine hydroxylase (TH) activity or dopamine (DA) metabolism. All three pretreatment (30 min) doses of desferal prevented the fall in striatal and frontal cortex DA, dihydroxyphenylacetic acid, and homovalinic acid, as well as the left and right striatum TH activity and DA turnover resulting from 6-OHDA lesion of dopaminergic neurons. A concentration bell-shaped neuroprotective effect of desferal was observed in the striatum, with 13 mug being the most effective. Neither desferal nor 6-OHDA affected striatal serotonin, 5-hydroxyindole acetic acid, or noradrenaline. Desferal also protected against 6-OHDA-induced deficit in locomotor activity, rearing, and exploratory behavior (sniffing) in a novel environment. Since the lowest neuroprotective dose (1.3 mug) of desferal was 200 times less than 6-OHDA, its neuroprotective activity may not be attributed to interference with the neurotoxin activity, but rather iron chelation. These studies led us to develop novel brain-permeable iron chelators, the VK-28 series, with iron chelating and neuroprotective activity similar to desferal for ironing iron out from PD and other neurodegenerative diseases, such as Alzheimer's disease, Friedreich's ataxia, and Huntington's disease.

Youdim MBH, Fridkin M, Zheng H. 2004. Novel bifunctional drugs targeting monoamine oxidase inhibition and iron chelation as an approach to neuroprotection in Parkinson's disease and other neurodegenerative diseases. J Neural Transm 111(10-11):1455-1471.
Abstract: Iron has been shown to accumulates at site where neurons degenerate in neurodegenerative diseases of Parkinson's disease, Alzheimer's disease, Huntington disease, amyotrophic lateral sclerosis and Friedreich ataxia. Iron is thought to participate or initiate oxidative stress via generation of reactive oxygen species (ROS), such as hydroxyl radical. Iron chelators are neuroprotective and prevent 6-hydroxydoapmine and MPTP dopaminergic neurotoxicity in rats and mice. However, their action on monoamine oxidase (MAO) A and B have not been determined previously since MAO-B inhibitors have been shown to be neuroprotective in cellular and animal models of Parkinson's disease. The chelators 8-hydroxyquinoline, O- phenanthroline, 2,2'-dipyridyl, U74500A and U74600F showed a preference for inhibition of rat brain mitochondrial MAO-A over MAO-B. Their IC50 ranged from 10(-3) M to 10(-6) M, with 21-amino steroids (U74500A and U74006F) showing a greater selectivity and potency for MAO-A. Desferrioxamine (desferal), a prototype potent iron chelator,

exhibited relatively poor MAO inhibitory. The inhibitions of MAO-A and B by 21-amino steroids (Lazaroids) were time dependent and irreversible. Those initiated by 8-hydroxyquinoline, 2,2;-dipyridyl and O-phenanthroline were fully reversible by enzyme dilution experiments. Both Fe2+ and Fe3+ reverse the MAO-A and B inhibition induced by the latter chelators, but not those initiated by 21-amino steroids. The data infer that either the inhibition of MAO by 21-amino steroids is either the resultant of their conversion to an irreversible covalently bound ligand or that the iron chelation moiety and MAO inhibitory activity in these compounds are not mutually shared. The results suggest that bifunctional brain penetrable drugs with iron chelating property and MAO inhibitory activity in could be the most feasible approach for neuroprotection in neurodegenerative diseases. Such drug would prevent participation of elevated iron in oxidative stress and formation of reactive hydroxyl radical, via its interaction with H(2)O2 (Fenton chemistry), generated as a consequence MAO and other oxidative enzyme reactions to generative cytotoxic reactive hydroxyl radical. We have now developed several of these compounds with neuroprotective, MAO inhibitory and iron chelating properties from our prototype iron chelators, VK-28 possessing propargylamine moiety of our anti-parkinson drug, rasagiline.

Yoshikawa K, Nakagawa M. 2004. Portal-systemic shunts, manganese, and parkinsonism - Reply. J Neurol Neurosurg Psychiatry 75(7):1081-1082.

Yen JH, Tsai WC, Lin CH, Ou TT, Hu CJ, Liu HW. 2004. Cytochrome P450 1A1 and manganese superoxide dismutase gene polymorphisms in Behcet's disease. J Rheumatol 31(4):736-740.
Abstract: Objective. To investigate the association of cytochrome p450 1A1 (CYP1A1) and manganese superoxide dismutase (MnSOD) gene polymorphisms with susceptibility to Behcet's disease (BD) in Taiwan. Methods. The polymorphisms of CYP1A1 and MnSOD genes were determined in 51 patients with BD and 91 healthy controls by polymerase chain reaction/restriction fragment length polymorphism methods. Results. The frequencies of CYP1A1 48896 and 4887A were significantly increased in patients with BD. In contrast, there was no significant difference in the frequencies of MnSOD gene polymorphisms between patients with BD and controls. Linkage disequilibrium was found between CYP1A1 48896 and CYP1A1 6235C in controls and patients with BD. However, a similar finding could not be found between CYP1A1 48896 and 4887A. We also found that the association of CYPIA1 48896 with BD was dependent on the presence of CYPIA1 4887A. On the other hand, the association of CYP1A1 4887A with BD was dependent on the presence of CYPIA1 48896. An additive effect of CYPIA1 48896 and 4887A on the susceptibility to BD could be found. Conclusion. Simultaneous presence of CYPIA1 48896 and 4887A is associated with development of BD in Taiwan.

Xu XY, Pin S, Gathinji M, Fuchs R, Harris ZL . 2004. Aceruloplasminemia - an Inherited Neurodegenerative Disease With Impairment of Iron HomeostasisVolume 1012. p 299-305. Redox-Active Metals in Neurological Disorders: Annals of the New York Academy of Sciences.
Abstract: In 1987, Miyajima et al. first characterized an autosomal recessive, adult-onset neurodegenerative disorder resembling Parkinson's disease associated with near-absent circulating serum ceruloplasmin levels. Coined "familial apoceruloplasmin deficiency", they described a patient with a presenting triad of diabetes mellitus, retinal degeneration, and neurodegeneration with blepharospasm. Neuropathological evaluation revealed abundant iron deposition in selected neurons of the basal ganglia and substantia nigra with associated neuronal dropout and spongioform degeneration without evidence of reactive gliosis. Subsequently, mutations in the ceruloplasmin gene have been determined to result in the excessive iron accumulation seen in the pancreas, retina, and brain. Elevated serum ferritin suggests a systemic iron overload syndrome, yet affected patients had low transferrin saturation and a mild anemia. This new disease, "aceruloplasminemia", reveals a role for ceruloplasmin as an essential ferroxidase critical for iron homeostasis. This multicopper oxidase

promotes efficient iron efflux such that individuals lacking ceruloplasmin develop a presumed oxidative injury secondary to iron accumulation and significant neuronal damage. Aceruloplasminemic mice provide a valuable model to further study the mechanisms by which ceruloplasmin regulates iron trafficking and the role of iron in oxidative injury. Despite the dependence of ceruloplasmin on copper for its function, aceruloplasminemia represents an iron storage disease and not a defect in copper metabolism. However, recent evidence in Saccharomyces cerevisiae indicates that Fet3, the yeast homologue of ceruloplasmin, functions as an essential cuprous oxidase. Further investigation into the mechanisms by which ceruloplasmin regulates iron and copper homeostasis will provide valuable insight into the pathogenesis of metallo-mediated diseases and elucidate mechanisms for transition metal (copper, iron) neuropathology.

Wissler JH. 2004. Extracellular and Circulating Redox- and Metalloregulated Erna and Ernp - Copper Ion-Structured Rna Cytokines (Angiotropin Ribokines) and Bioaptamer Targets Imparting Rna Chaperone and Novel Biofunctions to S100-Ef-Hand and Disease-Associated ProteinsVolume 1022. p 163-184. Circulating Nucleic Acids in Plasma/Serum Iii and Serum Proteomics: Annals of the New York Academy of Sciences.
Abstract: Bioassays for cellular differentiation and tissue morphogenesis were used to design methods for isolation of bioactive redox- and metalloregulated nucleic acids and copper ion complexes with proteins from extracellular, circulating, wound, and supernatant fluids of cultured cells. In extracellular biospheres, diversities of nucleic acids were found to be secreted by cells upon activation. They may reflect nucleic acid biolibraries with molecular imprints of cellular history. After removal of protein components, eRNA prototypes exuded by activated cells were sequenced. They are small, endogenous, highly modified and edited, redox- and metalloregulated 5'-end phosphorylated extracellular eRNA (similar to2-200 bases) with cellular, enzymic, and bioaptamer functions. Fenton-type OH* radical redox reactions may form modified nucleotides in RNA as wobbles eRNA per se, or as copper ion-complex with protein (e.g., S100A12-EF-hand protein, angiotropin-related protein, calgranulin-C, hippocampal neurite differentiation factor) are shown to be bioactive in vivo and in vitro as cytokines (ribokines) and as nonmitogenic angiomorphogens for endothelial cell differentiation in the formation of organoid supracellular capillary structures. As bioaptamers, copper ion-structured eRNA imparts novel biofunctions to proteins that they do not have on their own. The origin of extracellular RNA and intermediate precursors (up to 500 bases) was traced to intracellular parent nucleic acids. Intermediate precursors with and without partial homology were found. This suggests that bioaptamers are not directly retranslatable gene products. Metalloregulated eRNA bioaptamer function was investigated by domains (e.g. (5')...CUG...(3') hairpin loop) for folding, bioactivity, and binding of protein with copper, calcium, and alkali metal ion affinity. Vice versa, metalloregulated nucleic acid-binding domains (K3H, R3H) in proteins were identified. Interaction of protein and eRNA docking potentials were visualized by 3D-rapid prototyping of accurate molecular image models based on crystallographic or NMR data. For S100A12-homologous proteins, receptor- and metalloregulated RNA chaperone-shaped protein assemblies were investigated. They suggest insight into signaling cascades as to how eRNA transmits its cytokine (ribokine) bioinformation from the extracellular RNA biosphere into cells. Proteomics of the extracellular RNA biosphere demonstrate the presence of nucleic acid-binding domain homologies in defense-, aging-, and disease-associated neuronal and other proteins as targets for RNA orphans. By structural relationships found to transmissible processes, protein-aceous transfer ("infectivity") and feedback of bioinformation beyond the central dogma of molecular biology are considered in terms of metalloregulated RNA bioaptamer function, nucleic acid-binding domains, and protein conformation.

Windisch M, Hutter-Paier B, Schreiner E, Wronski R. 2004. beta-synuclein-derived peptides with neuroprotective activity - An alternative treatment of

neurodegenerative disorders? J Mol Neurosci 24(1):155-165.
Abstract: The 140-amino-acid protein alpha-synuclein (alpha-syn) is the major constituent of Lewy bodies. The protein interacts with several intracellular signal transduction pathways. Reasons for onset of abnormal aggregation of alpha-syn are unclear. Metal ions, oxidative stress, and beta-amyloid 1-42 (Abeta1-42) are important induction factors for alpha-syn aggregation. beta-Synuclein (beta-syn) can counteract alpha-syn aggregation. Cross-breeding of beta-syn transgenic mice with animals overexpressing alpha-syn significantly decreased alpha-syn-positive neuronal inclusion bodies and improved motor function. This was an important proof of concept for the role of beta-syn in regulating alpha-syn aggregation. A drug discovery program based on peptide derivatives (N-terminal amino acids 1-15) of beta-syn was initiated. For screening, tissue culture models simulating disease-specific conditions were utilized. They protected against growth factor withdrawal, Abeta toxicity, and oxidative stress. Three peptides were selected (KEGV, SMAKEGV, MDFMKGLSMAKE) for in vivo studies because they also decreased expression of Abeta1-40 and Abeta1-42. First, in vivo experiments were made in human amyloid precursor protein (APP [Swedish and London mutation]) transgenic mice, as well as a-syn transgenic mice. Treatment was performed with the peptides as an intraperitoneal injection or as intranasal droplets for 2 mo. Behavioral studies in APP transgenic mice were performed after 1 and 2 mo of treatment and showed clear effects of these peptides.

Williams E, Linert W. 2004. In vitro evidence supporting the therapeutic role of nicotine against neurodegeneration. In Vivo 18(3):391-399.
Abstract: This review supports the necessity of combining fundamental chemical and biological methods to scrutinize potential causative agents in neurodegeneration. This is supported by recent experimental evidence in relation to the use of nicotine as a potential therapeutic agent, especially when following the path of iron's role in catalysing the generation of reactive oxygen species via a Fenton like reaction. Exploration of the dose-response relationship indicates that acute administration offers the most likely success, reducing tremor and improving cognitive performance amongst others. Confirmation of this relationship is gathered from recent in vivo and in vitro efforts that support this hypothesis.

White AR, Barnham KJ, Huang X, Voltakis I, Beyreuther K, Masters CL, Cherny RA, Bush AI, Cappai R. 2004. Iron inhibits neurotoxicity induced by trace copper and biological reductants. Journal of Biological Inorganic Chemistry 9(3):269-280.
Abstract: The extracellular microenvironment of the brain contains numerous biological redox agents, including ascorbate, glutathione, cysteine and homocysteine. During ischemia/reperfusion, aging or neurological disease, extracellular levels of reductants can increase dramatically owing to dysregulated homeostasis. The extracellular concentrations of transition metals such as copper and iron are also substantially elevated during aging and in some neurodegenerative disorders. Increases in the extracellular redox capacity can potentially generate neurotoxic free radicals from reduction of Cu(II) or Fe(III), resulting in neuronal cell death. To investigate this in vitro, the effects of extracellular reductants (ascorbate, glutathione, cysteine, homocysteine or methionine) on primary cortical neurons was examined. All redox agents except methionine induced widespread neuronal oxidative stress and subsequent cell death at concentrations occurring in normal conditions or during neurological insults. This neurotoxicity was totally dependent on trace Cu (greater than or equal to0.4 muM) already present in the culture medium and did not require addition of exogenous Cu. Toxicity involved generation of Cu(I) and H2O2, while other trace metals did not induce toxicity. Surprisingly, administration of Fe(II) or Fe(III) (greater than or equal to2.5 muM) completely abrogated reductant-mediated neurotoxicity. The potent protective activity of Fe correlated with Fe inhibiting reductant-mediated Cu(I) and H2O2 generation in cell-free assays and reduced cellular Cu uptake by neurons. This demonstrates a novel role for Fe in blocking Cu-mediated neurotoxicity in a high reducing environment. A

possible pathogenic consequence for these phenomena was demonstrated by abrogation of Fe neuroprotection after pre-exposure of cultures to the Alzheimer's amyloid beta peptide (Abeta). The loss of Fe neuroprotection against reductant toxicity was greater after treatment with human Abeta1-42 than with human Abeta1-40 or rodent Abeta1-42, consistent with the central role of Abeta1-42 in Alzheimer's disease. These findings have important implications for trace biometal interactions and free radical-mediated damage during neurodegenerative illnesses such as Alzheimer's disease and old-age dementia.

Weinreb O, Mandel S, Amit T, Youdim MBH. 2004. Neurological mechanisms of green tea polyphenols in Alzheimer's and Parkinson's diseases. Journal of Nutritional Biochemistry 15(9):506-516.
Abstract: Tea consumption is varying its status from a mere ancient beverage and a lifestyle habit, to a nutrient endowed with possible prospective neurobiological-pharmacological actions beneficial to human health. Accumulating evidence suggest that oxidative stress resulting in reactive oxygen species generation and inflammation play a pivotal role in neurodegenerative diseases, supporting the implementation of radical scavengers, transition metal (e.g., iron and copper) chelators, and nonvitamin natural antioxidant polyphenols in the clinic. These observations are in line with the current view that polyphenolic dietary supplementation may have an impact on cognitive deficits in individuals of advanced age. As a consequence, green tea polyphenols are now being considered as therapeutic agents in well controlled epidemiological studies, aimed to alter brain aging processes and to serve as possible neuroprotective agents in progressive neurodegenerative disorders such as Parkinson's and Alzheimer's diseases. In particular, literature on the putative novel neuroprotective mechanism of the major green tea polyphenol, (-)-epigallocatechin-3-gallate, are examined and discussed in this review. (C) 2004 Elsevier Inc. All rights reserved.

Wanpen S, Govitrapong P, Shavali S, Sangchot P, Ebadi M. 2004. Salsolinol, a dopamine-derived tetrahydroisoquinoline, induces cell death by causing oxidative stress in dopaminergic SH-SY5Y cells, and the said effect is attenuated by metallothionein. Brain Res 1005(1-2):67-76.
Abstract: The endogenous neurotoxin, 1-methyl-6,7-dihydroxy-1,2,3,4-tetrahydroisoquinoline (salsolinol), has been considered a potential neurotoxin in the etiology of Parkinson's disease (PD). Salsolinol and N-methyl(R)-salsolinol were identified in the brains and cerebrospinal fluid (CSF) of PD patients. Oxidative stress is known to be one of the major contributing factors in the cascade that may finally leads to the cell death in PD. The present study was undertaken to understand the role of salsolinol in oxidative-mediated neuronal toxicity in dopaminergic SH-SY5Y cells, and the neuroprotective effects of metallothionein (MT) against salsolinol toxicity in MT overexpressing (MTtrans) fetal mesencephalic cells. Salsolinol increased the production of reactive oxygen species (ROS) and significantly decreased glutathione (GSH) levels and cell viability in SH-SY5Y cells. Salsolinol also decreased intracellular ATP levels and induced nuclear condensation in these cells. Salsolinol-induced depletion in cell viability was completely prevented by N-acetylcysteine in SH-SY5Y cells, and also prevented by MT in MTtrans fetal mesencephalic cells compared to control(wt) cells. The extent of nuclear condensation and caspase activation was also less in MTtrans cells than control(wt) cells. These results suggest that salsolinol causes oxidative stress by decreasing the levels of GSH and by increasing ROS production, and these events may lead to the death of dopaminergic cell. Furthermore, MT overexpression may protect dopaminergic neurons against salsolinol-induced neurotoxicity, most probably by the inhibition of oxidative stress and apoptotic pathways including caspase-3 activation. (C) 2004 Elsevier B.V All rights reserved.

Wang J, Jiang H, Xie JX. 2004. Time dependent effects of 6-OHDA lesions on iron level and neuronal loss in rat nigrostriatal system. Neurochem Res 29(12): 2239-2243.
Abstract: The early changes in iron level and neuronal loss in rat

nigrostriatal system were investigated using 6-hydroxydopamine (6-OHDA) unilaterally lesioned rats. The results showed that: 1, 3, 5, 7, and 14 days of postlesion, there was a progressive reduction in the density of the tyrosine hydroxylase immunoreactive (TH-ir) cells in the lesioned substantia nigra (SN). Iron level increased in the lesioned SN from 1 - 14 days following 6-OHDA lesions, but there were no differences in iron level among them. Only on 14 days of postlesion, did the DA release decrease in striatum (Str) of the lesioned side, while there were no changes in other groups. These results implied that the increased iron level in SN occured when there was a moderate reduction of DA neurons. However, the DA release in Str was unchanged until TH-ir cells were highly reduced due to the immense compensatory mechanism of the DA system.

Walter U, Dressler D, Benecke R. 2004. Brain parenchyma sonography for early and differential diagnosis of Parkinson's disease. Aktuelle Neurologie 31 (7):325-332.
Abstract: Brain parenchyma sonography is a non-invasive imaging technique extending the results of conventional imaging methods because of its different physical principle. In patients with idiopathic Parkinson's disease (IPD) or with parkinsonism due to parkin mutation a highly characteristic enlargement of the ultrasound signal (hyperechogenicity) of the substantia nigra (SN) can be detected that is already present in presymptomatic stages and could be used in future as a preclinical risk marker. SN hyperechogenicity is thought to be caused by increased amounts of iron bound to proteins other than ferritin. In patients with multiple-system atrophy (MSA) and progressive supranuclear palsy (PSP) typically a normoechogenic SN is found, whereas patients with corticobasal degeneration (CBD) and dementia with diffuse Lewy bodies (DLB) also exhibit SN hyperechogenicity. Clinical differentiation of IPD vs. MSA/PSP and of PSP vs. CBD, which is often difficult, is supported already in early stages by measurement of SN echogenicity. Further sonographic findings may improve syndrome discrimination. Abnormal lenticular-nucleus hyperechogenicity is present only in one-third of IPD and DLB patients, but in about 80% of MSA, PSP and CBD patients. Third-ventricle dilatation exceeding 10 mm is characteristic for PSP. Brain parenchyma sonography represents a new tool for early diagnosis and discrimination of parkinsonian disorders.

Vidal R, Ghetti B, Takao M, Brefel-Courbon C, Uro-Coste E, Glazier BS, Siani V, Benson MD, Calvas P, Miravalle L, Rascol O, Delisle MB. 2004. Intracellular ferritin accumulation in neural and extraneural tissue characterizes a neurodegenerative disease associated with a mutation in the ferritin light polypeptide gene. J Neuropathol Exp Neurol 63(4):363-380.
Abstract: Abnormal accumulation of ferritin was found to be associated with an autosomal dominant slowly progressing neurodegenerative disease clinically characterized by tremor, cerebellar ataxia, parkinsonism and pyramidal signs, behavioral disturbances. and cognitive decline. These symptoms may appear sequentially over a period of 4 decades. Pathologically, intranuclear and intracytoplasmic bodies were found in glia and subsets of neurons in the central nervous system as well as in extraneural tissue. Biochemical analyses of these bodies isolated from the striatum and cerebellar cortex revealed that ferritin light polypeptide (FTL) and ferritin heavy polypeptide (FTH1) were the main constituents. Molecular genetic studies revealed a 2-bp insertion mutation in exon 4 of the FTL gene. The resulting mutant polypeptide is predicted to have a carboxy terminus that is altered in amino-acid sequence and length. In tissue sections, the bodies were immunolabeled by anti-ferritin and anti-ubiquitin antibodies and were stained by Perls' method for ferric iron. Synthetic peptides homologous to the altered and wild-type carboxy termini were used to raise polyclonal antibodies. These novel antibodies as well as an antibody recognizing FTH1 immunolabeled the bodies. This study of this disorder has provided additional knowledge and insights in the growing area of ferritin-related neurodegeneration.

Verspohl EJ, Engfer A. 2004. Drug interactions in antiparkinsonian therapy.

Nervenheilkunde 23(3):151-+.
Abstract: Patients with parkinson symptoms are often co-medicated with drugs against additional diseases. Combination therapies may induce drug interactions including life-threatening events. Drug interactions cannot be anticipated on the basis of having a good pharmacological background but have to be confirmed using reviews or data banks. The relevant interactions of all ant parkinsonian drugs with themselves, with psychophamacological drugs, drugs used for gastrointestinal and cardiology complaints, with food, vitamins and iron salts, with drugs influencing bleeding, with narcotics and analgetics are described and listed. Hints to react on or correct the interactions are given. Thus drug safety would be increased.

Vatassery GT, Demaster EG, Lai JCK, Smith WE, Quach HT. 2004. Iron uncouples oxidative phosphorylation in brain mitochondria isolated from vitamin E-deficient rats. Biochimica Et Biophysica Acta-Molecular Basis of Disease 1688(3):265-273.
Abstract: Few, if any, studies have examined the effect of vitamin E deficiency on brain mitochondrial oxidative phosphorylation. The latter was studied using brain mitochondria isolated from control and vitamin E-deficient rats (13 months of deficiency) after exposure to iron, an inducer of oxidative stress. Mitochondria were treated with iron (2 to 50 muM) added as ferrous ammonium sulfate. Rates of state 3 and state 4 respiration, respiratory control ratios, and ADP/O ratios were not affected by vitamin E deficiency alone. However, iron uncoupled oxidative phosphorylation in vitamin E-deficient mitochondria, but not in controls. In vitamin E-deficient mitochondria, iron decreased ADP/O ratios and markedly stimulated state 4 respiration; iron had only a modest effect on these parameters in control mitochondria. Thus, vitamin E may have an important role in sustaining oxidative phosphorylation. Low concentrations of iron (2 to 5 muM) oxidized mitochondrial tocopherol that exists in two pools. The release of iron in brain may impair oxidative phosphorylation, which would be exacerbated by vitamin E deficiency. The results are important for understanding the pathogenesis of human brain disorders known to be associated with abnormalities in mitochondrial function as well as iron homeostasis (e.g., Parkinson's disease). (C) 2004 Elsevier B.V. All rights reserved.

Van Rensburg SJ, Berman P, Potocnik F, Macgregor P, Hon D, De Villiers N. 2004. 5-and 6-glycosylation of transferrin in patients with Alzheimer's disease. Metab Brain Dis 19(1-2):89-96.
Abstract: Transferrin is a glycosylated metal-carrying serum protein. One of the biological functions of glycosylation is to regulate the life span of proteins, less glycosylation leading to a faster clearance of a protein from the circulation. In the case of transferrin, this would indirectly also influence iron homeostasis. Higher glycosylation has been demonstrated in patients with Parkinson's disease and rheumatoid arthritis. A genetic variant of transferrin, TfC2, occurs with increased frequency in patients with Alzheimer's disease (AD), rheumatoid arthritis, and other diseases associated with a free radical etiology. Investigations have so far not revealed the reason for the pro-oxidative qualities of TfC2. In this study the glycosylation of Tf in AD (TfC1 homozygotes and TfC1C2 heterozygotes) was compared with alcohol-induced dementia (AID) patients and nondemented, age-matched controls, using isoelectric focusing followed by blotting with anti-Tf antibodies. In TfC1 homozygotes a shift was found toward higher sialylation, but in TfC1C2 heterozygotes the 5- and 6-sialylated bands were less concentrated. The decreased sialalytion found for TfC1C2 heterozygotes, may indicate that the pro-oxidative TfC2 molecules are removed from the circulation at a faster rate than TfC1. This may be of benefit to AD patients having TfC2, but still does not explain why this Tf variant is pro-oxidative.

Toimela T, Tahti H. 2004. Mitochondrial viability and apoptosis induced by aluminum, mercuric mercury and methylmercury in cell lines of neural origin. Arch Toxicol 78(10):565-574.

Abstract: Mercury and aluminum are considered to be neurotoxic metals, and they are often connected with the onset of neurodegenerative diseases. In this study, mercuric mercury, methylmercury and aluminum were studied in three different cell lines of neural origin. To evaluate the effects, mitochondrial cytotoxicity and apoptosis induced by the metals were measured after various incubation times. SH-SY5Y neuroblastoma, U 373MG glioblastoma, and RPE D407 retinal pigment epithelial cells were subcultured to appropriate cell culture plates and 0.01 - 1,000 muM concentrations of methylmercury, mercuric and aluminum chloride were added into the growth medium. In the assay measuring the mitochondrial dehydrogenase activity, WST-1, the cultures were exposed for 15 min, 24 or 48 h before measurement. Cells were allowed to recover from the exposure in part of the study. Apoptosis induced by the metals was measured after 6-, 24- and 48-h exposure times with the determination of activated caspase 3 enzyme. Mitochondrial assays showed a clear dose-response and exposure time-response to the metals. The most toxic was methylmercury (EC50 similar to 0.8 muM, 48 h), and the most sensitive cell line was the neuroblastoma cell line SH-SY5Y. Furthermore, there was marked mitochondrial activation, especially in connection with aluminum and methylmercury at low concentrations. This activation may be important during the initiation of cellular processes. All the metals tested induced apoptosis, but with a different time- course and cell-line specificity. In microscopic photographs, glioblastoma cells formed. brillary tangles, and neuroblastoma cells settled along the fibrilles in cocultures of glial and neuronal cell lines during aluminum exposure. The study emphasized the toxicity of methylmercury to neural cells and showed that aluminum alters various cellular activities.

Tings T, Schettler V, Canelo M, Paulus W, Trenkwalder C. 2004. Impact of regular LDL apheresis on the development of restless legs syndrome. Mov Disord 19(9):1072-1075.
Abstract: We examined 25 hyperlipidaemic patients with coronary heart disease undergoing regular low-density lipoprotein apheresis (LA) treatment in weekly intervals. In this patient population, half were found to have concomitant restless legs syndrome (RLS). Laboratory investigations suggest that iron metabolism is modified by regular LA treatment and this change may be involved in the pathogenesis of this previously unrecognised form of secondary RLS. Substitution of iron therefore may be a promising line of treatment for LA-induced RLS. (C) 2004 Movement Disorder Society.

Thomas M, Jankovic J. 2004. Neurodegenerative disease and iron storage in the brain. Curr Opin Neurol 17(4):437-442.
Abstract: Purpose of review Iron is very important for normal regulation of various metabolic pathways. Neurons store iron in the form of ferrous ion or neuromelanin. In specific disorders the axonal transport of iron is impaired, leading to iron deposition which in the presence of reactive oxygen species results in neurodegeneration. Recent findings Recent developments in genetics, including the finding of mutations in the pantothenate kinase gene and ferritin light chain gene, have demonstrated a direct relationship between the presence of a mutation in the iron-regulatory pathways and iron deposition in the brain resulting in neurodegeneration. These two disorders now add to our understanding of the mechanism of disease due to dysfunction of iron-regulatory pathways. In addition to these disorders there may be several other mutations of iron-regulatory genes or related genes that are yet to be found. The animal models of disease have also added value to this area. Summary In this review we provide a summary of recent developments in the field of movement disorders with abnormalities in iron transport, and the current evidence in neurodegenerative disorders such as Parkinson's disease.

Thomas M, Hayflick SJ, Jankovic J. 2004. Clinical heterogeneity of neurodegeneration with brain iron accumulation (Hallervorden-Spatz syndrome) and pantothenate kinase-associated neurodegeneration. Mov Disord 19(1):36-42.

Abstract: Hallervorden Spatz syndrome (HSS), also referred to as neurodegeneration with brain iron accumulation (NBIA), is a rare inherited neurodegenerative disorder with childhood, adolescent, or adult onset. Patients with HSS/NBIA have a combination of motor symptoms in the form of dystonia, parkinsonism, choreoathetosis, corticospinal tract involvement, optic atrophy, pigmentary retinopathy, and cognitive impairment. After the recent identification of mutations in the PANK2 gene on chromosome 20p12.3-p13 in some patients with the HSS/NBIA phenotype, the term pantothenate kinase-associated neurodegeneration (PKAN) has been proposed for this group of disorders. To characterize clinically and genetically HSS/ NBIA, we reviewed 34 affected individuals from 10 different families, who satisfied the inclusion criteria for NBIA. Relatives of patients who had clinical, magnetic resonance imaging (MRI), or pathological findings of NBIA were included in the study. Four patients were found to have mutations in the pantothenate kinase 2 (PANK2) gene. We compared the clinical features and MRI findings of those with and without PANK2 mutations. The presence of mutation in the PANK2 gene is associated with younger age at onset and a higher frequency of dystonia, dysarthria, intellectual impairment, and gait disturbance. Parkinsonism is seen predominantly in adult-onset patients whereas dystonia seems more frequent in the earlier-onset cases. The phenotypic heterogeneity observed in our patients supports the notion of genetic heterogeneity in the HSS/ NBIA syndrome. (C) 2003 Movement Disorder Society.

Takeda A. 2004. Essential trace metals and brain function. Yakugaku Zasshi 124 (9):577-585.
Abstract: Trace metals such as zinc, manganese, and iron are necessary for the growth and function of the brain. The transport of trace metals into the brain is strictly regulated by the brain barrier system, i.e., the blood-brain and blood-cerebrospinal fluid barriers. Trace metals usually serve the function of metalloproteins in neurons and glial cells, while a portion of trace metals exists in the presynaptic vesicles and may be released with neurotransmitters into the synaptic cleft. Zinc and manganese influence the concentration of neurotransmitters in the synaptic cleft, probably via the action against neurotransmitter receptors and transporters and ion channels. Zinc may be an inhibitory neuromodulator of glutamate release in the hippocampus, while neuromodulation by manganese might mean functional and toxic aspects in the synapse. Dietary zinc deficiency affects zinc homeostasis in the brain, followed by an enhanced susceptibility to the excitotoxicity of glutamate in the hippocampus. Transferrin may be involved in the physiological transport of iron and manganese into the brain and their utilization there. It is reported that the brain transferrin concentration is decreased in neurodegenerative diseases such as Alzheimer's disease and Parkinson's disease and that brain iron metabolism is also altered. The homeostasis of trace metals in the brain is important for brain function and also for the prevention of brain diseases.

Szczerbowska-Boruchowska M, Lankosz M, Ostachowicz J, Adamek D, Krygowska-Wajs A, Tomik B, Szczudlik A, Simionovici A, Bohic S. 2004. Topographic and quantitative microanalysis of human central nervous system tissue using synchrotron radiation. X-Ray Spectrometry 33(1): 3-11.
Abstract: Synchrotron microbeam X-ray fluorescence (mu-SXRF) was applied for topographic and quantitative analysis of selected elements in human brain and spinal cord. The main goal of the study is a better understanding of the role of elements, mainly metals, in processes leading to degeneration and atrophy of nerve cells in cases of two neurodegenerative disorders, i.e. Parkinson's disease (PD) and amyotrophic lateral sclerosis (ALS). The samples were taken during the autopsy from patients deceased with PD, ALS, and from a patient who died due to non-neurological conditions. In measurements, a 5 x 2 mum(2) X-ray beam was applied for scanning the thin tissue slices. Two-dimensional maps of elemental distribution were compared with histopathological sections. The results obtained showed that the spatial distribution of selected elements precisely corresponds to the microscopic views of

scanned area of tissue. Moreover, differences in accumulation of selected elements in nerve cell bodies and areas representing white matter between patients affected by PD, ALS and the control case were noticed. The present investigation showed that the mu-SXRF technique is suitable for elemental analysis at the single cell level in applications in neurological research. Copyright (C) 2004 John Wiley Sons, Ltd.

Sugimoto T, Ide-Ektessabi A, Ishihara R, Tanigaki A. 2004. An investigation of changes in element distribution and chemical states during differentiation of embryonic stem cells. Journal of Electron Spectroscopy and Related Phenomena 137-40(Sp. Iss. Si):831-838.
Abstract: Metallic elements and their organic compounds have dynamic regulatory functions in cells. In this study, we implemented a new approach to investigate the mechanism of differentiation of embryonic stem cells, by measuring and analyzing the change in distribution and chemical states of intracellular trace elements. We anticipate that trace metal elements and metalloproteins play important roles in the direction of differentiation, both as active centers, and as factors in the death of neural cells in neurodegenerative disorders. The aim of this study is to analyze the distribution and chemical states of trace elements during the process of differentiation of mouse embryonic stem cells, and to understand how these factors relate to the differentiation process. Using the experimental results, some previously unexplained points are considered, namely (1) how the intracellular elements change during the process of neuronal differentiation, and (2) what the optimal conditions of such elements are for neuronal differentiation. The information obtained during this study is relevant to nervous system development and evolution. (C) 2004 Elsevier B.V. All rights reserved.

Stroh A, Zimmer C, Gutzeit C, Jakstadt M, Marschinke F, Jung T, Pilgrimm H, Grune T. 2004. Iron oxide particles for molecular magnetic resonance imaging cause transient oxidative stress in rat macrophages. Free Radic Biol Med 36(8):976-984.
Abstract: Iron oxide particles are a promising marker in molecular magnetic resonance imaging. They are used to label distinct cell populations either in vitro or in vivo. We investigated for the first time whether small citrate-coated very small superparamagnetic iron oxide particles (VSOPs) can lead to an increase in cellular oxidative stress. We incubated rat macrophages (RAW) in vitro with iron oxide particles. We observed a massive uptake of VSOPs measured both with atomic absorption spectroscopy and with NMR, which could be visualized by confocal laser scanning microscopy. After incubation, cells were lysed and the levels of malonyldialdehyde (MDA) and protein carbonyls were determined. We found a significant increase in both MDA and protein carbonyl levels after incubation with the particles. Surprisingly, 24 h after incubation, a significant indication of oxidative stress could no longer be observed. The increase in oxidative stress seems to be transient and closely linked to the incubation procedure. The iron chelator desferal and the intracellular spin trap PBN caused a significant reduction in oxidative stress to almost control levels. This indicates that the augmentation of oxidative stress is closely linked to the free iron during incubation. Proliferation assays showed that incorporation of VSOPs did not lead to long-term cytotoxic effects even though the iron oxide particles remained in the cell. Magnetic labeling of cells with VSOPs seems to cause transient oxidative conditions not affecting cellular viability and seems to be a usable approach for molecular magnetic resonance imaging. (C) 2004 Elsevier Inc. All rights reserved.

Stredrick DL, Stokes AH, Worst TJ, Freeman WM, Johnson EA, Lash LH, Aschner M, Vrana KE. 2004. Manganese-induced cytotoxicity in dopamine-producing cells. Neurotoxicology 25(4):543-553.
Abstract: Manganese (Mn) is an essential metal that, at excessive levels in the brain, produces extrapyramidal symptoms similar to those in patients with Parkinson's disease (PD). In the present study, Mn toxicity was characterized in a human neuroblastoma (SK-N-SH) cell line and in a

mouse catecholaminergic (CATH.a) cell line. Mn was demonstrated to be more toxic in the catecholamine-producing CATH.a cells (EC50 = 60 muM) than in non-catecholaminergic SK-N-SH cells (EC50 = 200 muM). To test the hypothesis that the sensitivity of CATH.a cells to Mn is associated with their dopamine (DA) content, DA concentrations were suppressed in these cells by pretreatment with a-methyl-para-tyrosine (AMPT). Treatment for 24 h with 100 muM AMPT decreased intracellular DA, but offered no significant protection from Mn exposure (EC50 = 60 muM). Additional studies were carried out to assess if Mn toxicity was dependent on glutathione (GSH) levels. CATH.a cells were significantly protected by the addition of 5 mM GSH (Mn EC50 = 200 muM) and 10 mM N-acetyl cysteine (NAC) (Mn EC50 = 300 muM), therefore, indirectly identifying intracellular ROS formation as a mechanism for Mn neurotoxicity. Finally, apoptotic markers of Mn-induced cell death were investigated. DNA fragmentation, caspase-3 activation, and apoptosis-related gene expression were studied in CATH.a cells. No internucleosomal fragmentation or caspase activation was evident, even in the presence of supraphysiological Mn concentrations. cDNA hydridization array analysis with two differing Mn concentrations and time points, identified no noteworthy mRAA inductions of genes associated with programmed cell death. In conclusion, DA content was not responsible for the enhanced sensitivity of CATH.a cells to Mn toxicity, but oxidative stress was implicated as a probable mechanism of cytotoxicity. (C) 2003 Published by Elsevier Inc.

Sohmiya M, Tanaka M, Aihara Y, Okamoto K. 2004. Structural changes in the midbrain with aging and Parkinson's disease: an MRI study. Neurobiol Aging 25(4):449-453.
Abstract: We measured midbrain structures of 59 subjects with Parkinson's disease (PD) and 140 age- and gender-matched normal subjects without neurological disorders by using T2-weighted MR imaging. There is a significant increase in the maximum distance of the substantia nigra (SND) and a significant decrease in the average distance from the substantia nigra to the red nucleus (SNRND) in patients with PD compared with normal subjects in the 70 years old or less group. These findings may reflect the pathologic increase of iron concentration and the neuronal loss in the region. However, it is difficult to find differences between normal subjects and patients with PD in the greater than 70 years old group in the midbrain structures. These findings based on a large-scale morphometric study provide essential information to evaluate conventional MR images of PD. (C) 2003 Elsevier Inc. All rights reserved.

Snyder H, Wolozin B. 2004. Pathological proteins in Parkinson's disease: focus on the proteasome. J Mol Neurosci 24(3):425-442.
Abstract: Parkinson's disease (PD) is a multifactorial disease that appears to arise from the effects of both genetic and environmental influences. Pesticides and heavy metals are the principle environmental factors that appear to impact on PD. The known genetic factors include multiple genes that have been identified in related parkinsonian syndromes, as well as alpha-synuclein. Genes associated with either PD or Parkinson-related disorders include parkin, DJ-1, ubiquitin C-terminal hydrolase isozyme L1 (UCH-L1), nuclear receptor-related factor 1, and alpha-synuclein. alpha-Synuclein is particularly notable because it aggregates readily and is the main component of Lewy bodies (LBs). Aggregated alpha-synuclein binds the proteasome and potently inhibits proteasomal activity. Because ubiquitin accumulates in LBs, and parkin and UCH-L1 also interact with the ubiquitin proteasomal system, proteasomal dysfunction is thought to contribute to the pathophysiology of PD. Increasing numbers of experiments suggest that neurotoxins might interact with alpha-synuclein or other Parkinson-related proteins to contribute to the pathophysiology of PD. Transgenic animal models overexpressing alpha-synuclein develop age-dependent motor dysfunction and inclusions in the brain stem that contain alpha-synuclein. These models are very helpful in elucidating the pathophysiology of PD but do not completely recapitulate the disease process. The relationship between these transgenic models and PD is a

subject of intense investigation.

Singh SK, Grass G, Rensing C, Montfort WR. 2004. Cuprous oxidase activity of CueO from Escherichia coli. J Bacteriol 186(22):7815-7817.
Abstract: We have found CueO from Escherichia coli to have a robust cuprous oxidase activity, severalfold higher than any homologue. These data suggest that a functional role for CueO in protecting against copper toxicity in vivo includes the removal of Cu(I).

Simon D, Seznec H, Gansmuller A, Carelle N, Weber P, Metzger D, Rustin P, Koenig M, Puccio H. 2004. Friedreich ataxia mouse models with progressive cerebellar and sensory ataxia reveal autophagic neurodegeneration in dorsal root ganglia. J Neurosci 24(8):1987-1995.
Abstract: Friedreich ataxia (FRDA), the most common recessive ataxia, is characterized by degeneration of the large sensory neurons of the spinal cord and cardiomyopathy. It is caused by severely reduced levels of frataxin, a mitochondrial protein involved in iron-sulfur cluster (ISC) biosynthesis. Through a spatiotemporally controlled conditional gene-targeting approach, we have generated two mouse models for FRDA that specifically develop progressive mixed cerebellar and sensory ataxia, the most prominent neurological features of FRDA. Histological studies showed both spinal cord and dorsal root ganglia (DRG) anomalies with absence of motor neuropathy, a hallmark of the human disease. In addition, one line revealed a cerebellar granule cell loss, whereas both lines had Purkinje cell arborization defects. These lines represent the first FRDA models with a slowly progressive neurological degeneration. We identified an autophagic process as the causative pathological mechanism in the DRG, leading to removal of mitochondrial debris and apparition of lipofuscin deposits. These mice therefore represent excellent models for FRDA to unravel the pathological cascade and to test compounds that interfere with the degenerative process.

Shoham S, Youdim MBH. 2004. Nutritional Iron Deprivation Attenuates Kainate-Induced Neurotoxicity in Rats: Implications for Involvement of Iron in NeurodegenerationVolume 1012. p 94-114. Redox-Active Metals in Neurological Disorders: Annals of the New York Academy of Sciences.
Abstract: There is evidence suggesting that oxidative stress contributes to kainate neurotoxicity. Since iron promotes oxidative stress, the present study explores how change in nutritional iron content modulates kainate-induced neurotoxicity. Rats received an iron-deficient diet (ID) from 22 days of age for 4 weeks. One control group received the same diet supplemented with iron and another control group received standard rodent diet. Cellular damage after subcutaneous kainate (10 mg/kg) was assessed by silver impregnation and gliosis by staining microglia. ID reduced cellular damage in piriform and entorhinal cortex, in thalamus, and in hippocampal layers CA1-3. ID also attenuated gliosis, except in the hippocampal CA1 layer. Given involvement of zinc in hippocampal neurotransmission and in oxidative stress, we tested for a possible interaction of nutritional iron with nutritional zinc. Rats were made iron-deficient and then assigned to supplementation with iron, zinc, or iron + zinc. Controls were continued on ID diet. After 2 weeks, rats were treated with kainate. Iron supplementation abolished the protective effect of ID in piriform and entorhinal cortex. In hippocampal CA1 and dorsal thalamus, neither iron nor zinc supplementation alone abolished the protective effect of ID against cellular damage. Iron + zinc supplementation abolished ID protection in dorsal thalamus, but not in reuniens nucleus. Kainate-induced gliosis in CA1 remained unaffected by nutritional treatments. Thus, in piriform and entorhinal cortex, nutritional iron has a major impact on cellular damage and gliosis. In hippocampal CA1, gliosis may associate with synaptic plasticity not modulated by nutritional iron, while cellular damage is Sensitive to nutritional iron and zinc.

Sharma SK, Ebadi M. 2004. An improved method for analyzing coenzyme Q homologues and multiple detection of rare biological samples. J Neurosci Methods 137(1):1-8.

Abstract: We have developed a simple method for the estimation of coenzyme Q homologues, neurotransmitters, metal ions, lipid peroxidation, gene expression, and DNA fragmentation simultaneously from genetically engineered mice brain regions and cultured neurons. The primary objective of this study was to improve conventional time-consuming, cumbersome, and less efficient procedures, and reduce the cost of conducting kinetic studies in rare biological samples. The improved method is novel, precise, efficient, accurate, sensitive, economical, versatile, and highly reproducible. The recovery and shelf life of coenzyme Q homologues was significantly increased and the chromatograms exhibited reduced background and retention times. It is envisaged that in addition to coenzyme Q homologues, the improved method could be utilized for the multiple analyses of DNA, RNA and proteins from clinically significant biopsy and autopsy samples. (C) 2004 Elsevier B.V. All rights reserved.

Shang T, Kotamraju S, Kalivendi SV, Hillard CJ, Kalyanaraman B. 2004. 1-methyl-4-phenylpyridinium-induced apoptosis in cerebellar granule neurons is mediated by transferrin receptor iron-dependent depletion of tetrahydrobiopterin and neuronal nitric-oxide synthase-derived superoxide. J Biol Chem 279(18):19099-19112.
Abstract: In this study, we investigated the molecular mechanisms of toxicity of 1-methyl-4-phenylpyridinium (MPP+), an ultimate toxic metabolite of a mitochondrial neurotoxin, 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine, that causes Parkinson-like symptoms in experimental animals and humans. We used rat cerebellar granule neurons as a model cell system for investigating MPP+ toxicity. Results show that MPP+ treatment resulted in the generation of reactive oxygen species from inhibition of complex I of the mitochondrial respiratory chain, and inactivation of aconitase. This, in turn, stimulated transferrin receptor (TfR)-dependent iron signaling via activation of the iron-regulatory protein/iron-responsive element interaction. MPP+ caused a time-dependent depletion of tetrahydrobiopterin (BH4) that was mediated by H2O2 and transferrin iron. Depletion of BH4 decreased the active, dimeric form of neuronal nitric-oxide synthase (nNOS). MPP+-mediated "uncoupling" of nNOS decreased .NO and increased superoxide formation. Pretreatment of cells with sepiapterin to promote BH4 biosynthesis or cell-permeable iron chelator and TfR antibody to prevent iron-catalyzed BH4 decomposition inhibited MPP+ cytotoxicity. Preincubation of cerebellar granule neurons with nNOS inhibitor exacerbated MPP+-induced iron uptake, BH4 depletion, proteasomal inactivation, and apoptosis. We conclude that MPP+-dependent aconitase inactivation, Tf-iron uptake, and oxidant generation result in the depletion of intracellular BH4, leading to the uncoupling of nNOS activity. This further exacerbates reactive oxygen species-mediated oxidative damage and apoptosis. Implications of these results in unraveling the molecular mechanisms of neurodegenerative diseases (Parkinson's and Alzheimer's disease) are discussed.

Sevigny JJ, Albert SM, Mcdermott MP, Mcarthur JC, Sacktor N, Conant K, Schifitto G, Selnes OA, Stern Y, Mcclernon DR, Palumbo D, Kieburtz K, Riggs G, Cohen B, Epstein LG, Marder K. 2004. Evaluation of HIV RNA and markers of immune activation as predictors of HIV-associated dementia. Neurology 63(11):2084-2090.
Abstract: Objective: To evaluate whether baseline levels of plasma and CSF HIV RNA, tumor necrosis factor alpha (TNFalpha), monocyte chemoattractant protein-1 (MCP-1), matrix metalloproteinase-2 (MMP-2), or macrophage colony stimulating factor (M-CSF) are predictors of incident HIV-associated dementia (HIVD) in a cohort with advanced HIV infection. Methods: A total of 203 nondemented subjects with CD4 lymphocyte counts less than 200/muL, or <300/μL but with cognitive impairment, underwent semiannual neurologic, cognitive, functional, and laboratory assessments. HIVD and minor cognitive motor disorder (MCMD) were defined using American Academy of Neurology criteria. The cumulative incidence of HIVD was estimated using Kaplan-Meier curves. Cox proportional hazards regression models were used to examine the associations between biologic variables and time to HIVD, adjusting for

age, sex, years of education, duration of HIV infection, type of antiretroviral use, premorbid IQ score, and presence of MCMD. Results: After a median follow-up time of 20.7 months, 74 (36%) subjects reached the HIVD endpoint. The dementia was mild in 70% of cases. The cumulative incidence of HIVD was 20% at 1 year and 33% at 2 years. Highly active antiretroviral therapy (HAART) was used by 73% of subjects at baseline. A plasma HIV RNA level was undetectable in 23% of subjects and a CSF HIV RNA level was undetectable in 48% of subjects. In adjusted analyses, neither plasma nor CSF HIV RNA levels (log(10)) were associated with time to HIVD; log10 levels of plasma TNFα (HR 3.07, p = 0.03) and CSF MCP-1 (HR = 3.36, p = 0.06) tended to be associated with time to HIVD. Conclusion: The lack of association between baseline plasma and CSF HIV RNA levels and incident dementia suggests highly active antiretroviral therapy may be affecting CNS viral dynamics, leading to lower HIV RNA levels, and therefore weakening the utility of baseline HIV RNA levels as predictors of HIV-associated dementia.

Schipper HM. 2004. Heme oxygenase expression in human central nervous system disorders. Free Radic Biol Med 37(12):1995-2011.
Abstract: In the normal mammalian CNS, heme oxygenase-2 (HO-2) is constitutively, abundantly, and fairly ubiquitously expressed, whereas heme oxygenase-1 (HO-1) mRNA and protein are confined to small populations of scattered neurons and neuroglia. Unlike ho-2, the ho-1 gene in neural (and many systemic) tissues is exquisitely sensitive to Upregulation by a host of pro-oxidant and other noxious stimuli. In Alzheimer disease, HO-1 immunoreactivity is significantly augmented in neurons and astrocytes of the hippocampus and cerebral cortex relative to age-matched, nondemented controls and colocalizes to senile plaques, neurofibrillary tangles, and corpora amylacea. In Parkinson disease, HO-I decorates Lewy bodies of affected dopaminergic neurons and is highly overexpressed in astrocytes residing within the substantia nigra. The ho-1 gene is also upregulated in glial cells within multiple sclerosis plaques; in the vicinity of human cerebral infarcts, hemorrhages, and contusions; and in various other degenerative and non degenerative human CNS disorders. The products of the heme oxygenase reaction, free ferrous iron, carbon monoxide, and biliverdin/bilirubin, are all biologically active molecules that may profoundly influence tissue redox homeostasis under a wide range of pathophysiological conditions. Evidence adduced from whole animal and in vitro studies indicates that enhanced HO-1 activity may either ameliorate or exacerbate neural injury, effects likely contingent upon the specific model employed, the duration and intensity of HO-I induction, and the chemistry of the local redox microenvironment. HO-1 hyperactivity also promotes mitochondrial sequestration of nontransferrin iron in oxidatively challenged astroglia and may thereby contribute to the pathological iron deposition and bioenergetic failure amply documented in aging and degenerating human neural tissues. (C) 2004 Elsevier Inc. All rights reserved.

Schipper HM. 2004. Heme Oxygenase-1: Transducer of Pathological Brain Iron Sequestration Under Oxidative StressVolume 1012. p 84-93. Redox-Active Metals in Neurological Disorders: Annals of the New York Academy of Sciences.
Abstract: Mechanisms responsible for the pathological deposition of redox-active brain iron in human neurological disorders remain incompletely understood. Heme oxygenase-1 (HO-1) is a 32-kDa stress protein that degrades heme to biliverdin, free iron, and carbon monoxide. In this; chapter, we review evidence that (1) HO-1 is overexpressed in CNS tissues affected by Alzheimer's disease (AD), Parkinson's disease (PD), multiple sclerosis (MS), and other degenerative and nondegenerative CNS diseases; (2) the pro-oxidant effects of dopamine, hydrogen peroxide, beta-amyloid, and proinflammatory cytokines stimulate HO-1 expression in some of these conditions; and (3) upregulation of HO-1 in astrocytes exacerbates intracellular oxidative stress and promotes sequestration of nontransferrin-derived iron by the mitochondrial compartment. A model is presented implicating glial HO-1 induction as a "final common pathway"

leading to pathological iron sequestration and mitochondrial insufficiency in a host of human CNS disorders.

Schiewe AJ, Margol L, Soreghan BA, Thomas SN, Yang AJ. 2004. Rapid characterization of amyloid-beta side-chain oxidation by tandem mass spectrometry and the scoring algorithm for spectral analysis. Pharm Res 21(7):1094-1102.
Abstract: Purpose. Amyloid-beta (Abeta) is a self-aggregating protein found in senile plaques in Alzheimer's disease (AD) brain and is thought to play a major role in the disease process. Oxidative stress may be a predominant cause of the formation of these Abeta aggregates. This study aims at identifying possible sites of copper-catalyzed oxidation of Abeta1-40 using liquid chromatography tandem mass spectrometry (LC/MS/MS) and scoring algorithm for spectral analysis (SALSA). Traditionally, identification of post-translational modifications by tandem mass spectrometric analysis requires users to inspect manually thousands of MS/MS spectra, which can be a tedious and time-consuming process. With the use of SALSA, users can automatically search for post-translational modifications based on the spacing of the m/z values associated with the ion series of an amino acid sequence. Methods. Abeta1-40 was subjected to copper-catalyzed oxidative stress. LC/MS/MS and SALSA analyses were used to determine the sites of post-translational modification within the tryptic fragments. Results. Oxidation was found to occur preferentially at the histidine residues His13 and His14 and at the methionine residue (Met35) of Abeta1-40. Conclusions. The combination of LC/MS/MS and SALSA searches could dramatically improve the efficiency and accuracy of determining the specific sites of oxidation of in vitro, copper-oxidized Abeta1-40 as well as other oxidized proteins.

Schenck JF, Zimmerman EA. 2004. High-field magnetic resonance imaging of brain iron: birth of a biomarker? NMR Biomed 17(7):433-445.
Abstract: The brain has an unusually high concentration of iron, which is distributed in an unusual pattern unlike that in any other organ. The physiological role of this iron and the reasons for this pattern of distribution are not yet understood. There is increasing evidence that several neurodegenerative diseases are associated with altered brain iron metabolism. Understanding these dysmetabolic conditions may provide important information for their diagnosis and treatment. For many years the iron distribution in the human brain could be studied effectively only under postmortem conditions. This situation was changed dramatically by the finding that T-2-weighted MR imaging at high field strength (initially 1.5 T) appears to demonstrate the pattern of iron distribution in normal brains and that this imaging technique can detect changes in brain iron concentrations associated with disease states. Up to the present time this imaging capability has been utilized in many research applications but it has not yet been widely applied in the routine diagnosis and management of neurodegenerative disorders. However, recent advances in the basic science of brain iron metabolism, the clinical understanding of neurodegenerative diseases and in MRI technology, particularly in the availability of clinical scanners operating at the higher field strength of 3 T, suggest that iron-dependent MR imaging may soon provide biomarkers capable of characterizing the presence and progression of important neurological disorders. Such biomarkers may be of crucial assistance in the development and utilization of effective new therapies for Alzheimer's and Parkinson's diseases, multiple sclerosis and other iron-related CNS disorders which are difficult to diagnose and treat. Copyright (C) 2004 John Wiley Sons, Ltd.

Sava V, Mosquera D, Song SJ, Cardozo-Pelaez F, Sanchez-Ramos JR. 2004. Effects of melanin and manganese on DNA damage and repair in PC 12-derived neurons. Free Radic Biol Med 36(9):1144-1154.
Abstract: The mechanism of neurotoxicity produced by the interaction of melanin with manganese was investigated in PC12-derived neuronal cell cultures. The cells were incubated with melanin (25-500 mug/ml), MnCl2 (10 ng/ml-100 mug/ ml), and a combination of both substances for 24 and

72 h. Incubation with either toxicant alone resulted in a minimal decrease in cell viability. The combination of melanin and manganese caused significant (up to 60%) decreases in viability of PC12 cells in a dose-dependent manner. Increases in oxidative DNA damage, indicated by levels of 8-hydroxy-2'deoxyguanosine (8-oxodG), was associated with decreased cell viability. Melanin alone, but not manganese alone, resulted in increased oxidative DNA damage. The maximal increase in 8-oxodG caused by melanin was about seven times higher than control after 24 h of exposure. The activity of the DNA repair enzyme, 8-oxoguanine DNA glycosylase (OGG1), was increased in cells incubated with single toxicants and their combinations for 24 h. On the third day of incubation with the toxicants, activity of OGG1 declined below control levels and cell viability significantly decreased. Melanin was observed to have an inhibitory effect on OGG1 activity. Study of the regulation of OGG1 activity in response to melanin and manganese may provide insights into the vulnerability of nigral neurons to oxidative stress in Parkinson's disease. (C) 2004 Elsevier Inc. All rights reserved.

Saarela MS, Lehtimaki T, Rinne JO, Hervonen A, Jylha M, Roytta M, Ahonen JP, Mattila KM. 2004. Interaction between matrix metalloproteinase 3 and the epsilon 4 allele of apolipoprotein E increases the risk of Alzheimer's disease in Finns. Neurosci Lett 367(3):336-339.
Abstract: Polymorphisms affecting the expression of matrix metalloproteinases (MMPs), i.e. proteolytic enzymes that degrade intercellular material, have been found at position -1607 (1G/2G) in MMP1 and at -1171 (5A/6A) in MMP3. Interestingly, elevated levels of MMP1 and MMP3 have been observed in the brains of Alzheimer's disease (AD) patients and those of tissue inhibitors of MMPs in the cerebrospinal fluid of AD and Parkinson's disease (PD) patients, suggesting a role for NIMPs in these disorders. The aim was to investigate a possible association between the afore-mentioned MMP1 and MMP3 polymorphisms and the risk of developing AD or PD. The polymorphisms were genotyped in 97 AD, 52 PD and 101 control patients. We found an interaction between MMP3*5A and APOE epsilon4 alleles ($P < 0.0001$) which increases the risk of AD (OR: 23.7, 95% CI: 5.8-144.9, $P < 0.0001$) compared to those who possess neither MMP3*5A nor APOE epsilon4. In conclusion, our finding suggests that the MMP3 gene, especially together with APOE epsilon4, may contribute to the development of AD. (C) 2004 Elsevier Ireland Ltd. All rights reserved.

Rossi L, Lombardo MF, Ciriolo MR, Rotilio G. 2004. Mitochondrial dysfunction in neurodegenerative diseases associated with copper imbalance. Neurochem Res 29(3):493-504.
Abstract: Copper is an essential transition metal ion for the function of key metabolic enzymes, but its uncontrolled redox reactivity is source of reactive oxygen species. Therefore a network of transporters strictly controls the trafficking of copper in living systems. Deficit, excess, or aberrant coordination of copper are conditions that may be detrimental, especially for neuronal cells, which are particularly sensitive to oxidative stress. Indeed, the genetic disturbances of copper homeostasis, Menkes' and Wilson's diseases, are associated with neurodegeneration. Furthermore, copper interacts with the proteins that are the hallmarks of neurodegenerative disorders, such as Alzheimer's disease, Parkinson's disease, prion diseases, and familial amyotrophic lateral sclerosis. In all cases, copper-mediated oxidative stress is linked to mitochondrial dysfunction, which is a common feature of neurodegeneration. In particular we recently demonstrated that in copper deficiency, mitochondrial function is impaired due to decreased activity of cytochrome c oxidase, leading to production of reactive oxygen species, which in turn triggers mitochondria-mediated apoptotic neurodegeneration.

Ritchie CW, Bush AI, Masters CL. 2004. Metal-protein attenuating compounds and Alzheimer"s disease. Expert Opinion on Investigational Drugs 13(12): 1585-1592.
Abstract: Since the description of the amyloid plaque in the pathology of

Alzheimer's disease, one of the main focuses of research has been the role of the amyloid precursor protein metabolite amyloid-P, which is the constituent protein of plaque. Affecting the production, aggregation or clearance of this protein may well have a modifying effect on disease progression. Although available therapies for Alzheimer's disease may interact with amyloid-P in vivo, no conspicuous disease-modifying effect has been demonstrated in clinical trials with these drugs. Drugs whose primary target is not the rectification of the neurotransmitter deficits associated with Alzheimer's disease but rather the life cycle of amyloid-beta are currently being developed with varying degrees of success. Of these drugs, the metal-protein attenuating compounds have currently the most encouraging clinical data supporting their use. Clioquinol is an example from this class, which has recently shown encouraging efficacy from early clinical evaluation in the absence of any compelling evidence of subacute myelopathic optic neuritis, which has been associated with this drug's use in Japanese populations. This article will discuss the scientific rationale behind the use of metal-protein attenuating compounds in Alzheimer's disease and summarise the available clinical trial data.

Riederer PF. 2004. Views on neurodegeneration as a basis for neuroprotective strategies. Medical Science Monitor 10(12):RA287-RA290.
Abstract: Evidence is presented to demonstrate neurodegenerative processes in Parkinson's disease which are interconnected and may be synergistic in a way that they self-perpetuate progression. Free iron plays a predominant role, because it may be continuously and unlimitedly taken up through a disturbed blood-brain-barrier. Iron's toxic action is at both neuronal and glial sites. Loss of tyrosine hydroxylase protein and activity and fibrillation of a-synuclein connected with disturbed proteasomal protein breakdown contribute to cell death, as are changes in neuromelanin concentration and binding affinity, e.g. for iron. The interplay of genetic disturbances and neuronal and glial pathological processes involving the functioning of the blood-brain barrier, eventually initiated via an ascending toxic process, is the key for attacking vulnerable catecholaminergic neurons such as those in the substantia nigra and locus coeruleus. Neuroprotective therapeutic strategies are difficult to achieve because of the immanent complexity of cell death cascades.

Richardson DR. 2004. Novel Chelators for Central Nervous System Disorders That Involve Alterations in the Metabolism of Iron and Other Metal IonsVolume 1012. p 326-341. Redox-Active Metals in Neurological Disorders: Annals of the New York Academy of Sciences.
Abstract: Recent evidence suggests that iron (Fe) and other metals play a role in a number of neurodegenerative diseases including: Friedreich's ataxia, Alzheimer's disease, Huntington's disease, and Parkinson's disease. In this review, the role of Fe and other metals in the pathology of these conditions is assessed and the potential of Fe chelators for treatment is discussed. Lipophilic chelators have been designed that may be capable of crossing the blood-brain barrier, a property lacking in desferrioxamine (DFO), a chelator in widespread clinical use. A far less commonly used chelator, clioquinol, has already shown activity in vivo in animal models and also in Alzheimer's disease patients. Considering that there is no effective treatment for many neurological diseases, the therapeutic use of lipophilic Fe chelators remains a potential strategy that requires investigation. In particular, we discuss the development of several series of aroylhydrazone chelators that could have high potential in the treatment of these diseases.

Rao KVR, Norenberg MD. 2004. Manganese induces the mitochondrial permeability transition in cultured astrocytes. J Biol Chem 279(31): 32333-32338.
Abstract: Manganese is known to cause central nervous system injury leading to parkinsonism and to contribute to the pathogenesis of hepatic encephalopathy. Although mechanisms of manganese neurotoxicity are not completely understood, chronic exposure of various cell types to manganese has shown oxidative stress and mitochondrial energy failure,

factors that are often implicated in the induction of the mitochondrial permeability transition (MPT). In this study, we examined whether exposure of cultured neurons and astrocytes to manganese induces the MPT. Cells were treated with manganese acetate (10-100 muM), and the MPT was assessed by changes in the mitochondrial membrane potential and in mitochondrial calcein fluorescence. In astrocytes, manganese caused a dissipation of the mitochondrial membrane potential and decreased the mitochondrial calcein fluorescence in a concentration- and time-dependent manner. These changes were completely blocked by pretreatment with cyclosporin A, consistent with induction of the MPT. On the other hand, similarly treated cultured cortical neurons had a delayed or reduced MPT as compared with astrocytes. The manganese-induced MPT in astrocytes was blocked by pretreatment with antioxidants, suggesting the potential involvement of oxidative stress in this process. Induction of the MPT by manganese and associated mitochondrial dysfunction in astrocytes may represent key mechanisms in manganese neurotoxicity.

Purdey M. 2004. Elevated levels of ferrimagnetic metals in foodchains supporting the Guam cluster of neurodegeneration: Do metal nucleated crystal contaminents evoke magnetic fields that initiate the progressive pathogenesis of neurodegeneration? Med Hypotheses 63(5):793-809. Abstract: Elevated levels of aluminium (At), strontium (Sr), barium (Ba), iron (Fe), manganese (Mn) cations combined with deficiencies of magnesium (Mg)/calcium (Ca) - have been observed in the foodchains that traditionally support the Chamorro populations affected by high incidence clusters of Alzheimer (AD), Parkinson-like (PD), motor neurone diseases and multiple sclerosis on the island of Guam. Soils drawn from the cluster region demonstrated an excessive fivefold increase in 'magnetic susceptibility' readings in relation to soils from disease free adjoining regions. A multifactorial aetiological hypothesis is proposed that pivots upon the combined exposure to high levels of natural/industrial sources of ferrimagnetic/ferroelectric compounds incorporating A[, Fe, Mn, Sr, Ba (e.g., via yam/seafood consumption or exposure to world war 2 (WW2) munitions) and to low levels of Mg/Ca in all S. Pacific locations where these clusters of neurodegenerative disease have simultaneously erupted. Once gut/blood brain barrier permeability is impaired, the increased uptake of At, Fe, Sr, Ba, or Mn into the Mg/Ca depleted brain Leads to rogue metal substitutions at the Mg/Ca vacated binding domains on various enzyme/ proteoglycan groups, causing a broad ranging disruption in Mg/Ca dependent systems - such as the glutamine synthetase which prevents the accumulation of neurotoxic glutamate. The rogue metals chelate sulphate, disrupting sulphated-proteoglycan mediated inhibition of crystal proliferation, as well as its regulation of the Fibroblast growth factor receptor complex which disturbs the molecular conformation of those receptors and their regulation of transphosphorylation between intracellular kinase domains; ultimately collapsing proteoglycan mediated cell-cell signalling pathways which maintain the growth and structural integrity of the neuronal networks. The depression of Mg/Ca dependent systems in conjunction with the progressive ferrimagnetisation of the CNS due to an overload of rogue ferroelectric/ferrimagnetic metal contaminants, enables 'seeding' of metal-protein crystalline arrays that can proliferate in the proteoglycan depleted brain. The resulting magnetic field emissions initiate a free radical mediated progressive pathogenesis of neurodegeneration. The coclustering of these various types of disease in select geographical pockets around the world suggests that all of these conditions share a common early life exposure to ferromagnetic metal nucleating agents in their multifactorial aetiology. Factors such as individual genetics, the species of metal involved, etc., dictate which specific class of disease will emerge as a delayed neurotoxic response to these environmental insults. (C) 2004 Elsevier Ltd. All rights reserved.

Pifl C, Khorchide M, Kattinger A, Reither H, Hardy J, Hornykiewicz O. 2004. alpha-Synuclein selectively increases manganese-induced viability loss in SK-N-MC neuroblastoma cells expressing the human dopamine transporter. Neurosci Lett 354(1):34-37.

Abstract: The established or potentially toxic agents implicated in the nigral cell death in Parkinson's disease, dopamine, 1-methyl-4-phenylpyridinium (MPP+), iron, and manganese, were examined as to their effects on the viability of cells overexpressing alpha-synuclein. SKN-MC neuroblastoma cells stably expressing the human dopamine transporter were transfected with human alpha-synuclein and cell clones with and without alpha-synuclein immunoreactivity were obtained. Cells were exposed for 24-72 h to 1-10 muM dopamine, 0.1-3 muM MPP+, 0.1-1 mM FeCl2 or 30-300 muM MnCl2 added to the culture medium. There was no difference between cells expressing alpha-synuclein and control cells after exposure to dopamine, MPP+ or FeCl2. However, MnCl2 resulted in a significantly stronger decreased viability of cells overexpressing alpha-synuclein after 72 h. These findings suggest that manganese may co-operate with alpha-synuclein in triggering neuronal cell death such as seen in manganese parkinsonism. The relevance of our observations for the pathoetiology of Parkinson's disease proper remains to be determined. (C) 2003 Elsevier Ireland Ltd. All rights reserved.

Peng J, Mao XO, Stevenson FF, Hsu M, Andersen JK. 2004. The herbicide paraquat induces dopaminergic nigral apoptosis through sustained activation of the JNK pathway. J Biol Chem 279(31):32626-32632.
Abstract: Environmental exposure to the oxidant-producing herbicide paraquat has been implicated as a risk factor in Parkinson's disease. Although intraperitoneal paraquat injections in mice cause a selective loss of dopaminergic neurons in the substantia nigra pars compacta, the exact mechanism involved is still poorly understood. Our data show that paraquat induces the sequential phosphorylation of c-Jun N-terminal kinase (JNK) and c-Jun and the activation of caspase-3 and sequential neuronal death both in vitro and in vivo. These effects are diminished by the specific JNK inhibitor SP600125 and the antioxidant manganese(III) tetrakis (4-benzoic acid) porphyrin in vitro. Furthermore, JNK pathway inhibitor CEP-11004 effectively blocks paraquat-induced dopaminergic neuronal death in vivo. These results suggest that the JNK signaling cascade is a direct activator of the paraquat-mediated nigral dopaminergic neuronal apoptotic machinery and provides a molecular linkage between oxidative stress and neuronal apoptosis.

Park J, Yo CI, Simi CS, Kim JW, Yi YJ, Jung KY, Chung SE, Kim Y. 2004. Occupations and Parkinson's disease: A case-control study in south Korea. Ind Health 42(3):352-358.
Abstract: We performed a hospital based case-control study in the southeast region of Korea to clarify the role of occupational exposure, especially manganese (Mn), in the etiology of Parkinson's disease (PD) and to discover the association between any occupation and PD. 105 outpatients with PD and 129 neurological disease controls and 101 healthy controls were interviewed. We employed occupational and industrial categories as defined by Section (the most broad category) and Division (sub-category) of the Korea Standard Industry Code and the Korea Standard Classification of Occupations. There was not a significant association between exposure to hazardous materials, especially Mn and PD. There were not any occupations listed under the Section of Industry Classification as a significant risk factor or protective factor for PD. However, the 'clerk' occupation [Section] was positively associated with PD. There is a decreased risk for PD with a subject ever having worked in the 'agriculture, forestry and fishery' occupational group. Ever having worked in 'sales' also was negatively associated with PD. There were not any Divisions of Industry found as a significant risk factor or protective factor for PD. However, ever having worked in an 'agriculture' Division of Occupation was negatively associated with PD.

Olanow CW. 2004. Manganese-Induced Parkinsonism and Parkinson's DiseaseVolume 1012. p 209-223. Redox-Active Metals in Neurological Disorders: Annals of the New York Academy of Sciences.
Abstract: It has long been appreciated that manganese exposure can cause neurotoxicity and a neurologic syndrome that resembles Parkinson's

disease (PD). Current evidence indicates that manganese-induced parkinsonism can be differentiated from PD because of its predilection to accumulate in and damage the pallidum and striatum rather than the SNc. The clinical syndrome, response to levodopa, imaging studies with MRI and PET, and pathologic features all help to distinguish these two conditions and permit the correct diagnosis to be established. This is of particular relevance in differentiating patients with parkinsonism due to manganese intoxication from patients with idiopathic PD who have incidental manganese exposure.

Nunez MT, Gallardo V, Munoz P, Tapia V, Esparza A, Salazar J, Speisky H. 2004. Progressive iron accumulation induces a biphasic change in the glutathione content of neuroblastoma cells. Free Radic Biol Med 37(7):953-960. Abstract: Glutathione (GSH) constitutes the single most important antioxidant in neurons, whereas iron causes oxidative stress that leads to cell damage and death. Although GSH and iron produce opposite effects on redox cell status, no mechanistic relationships between iron and GSH metabolism are known. In this work, we evaluated in SHSY5Y neuroblastoma cells the effects of iron accumulation on intracellular GSH metabolism. After 2 d exposure to increasing concentrations of iron, cells underwent concentration-dependent iron accumulation and a biphasic change in intracellular GSH levels. Increasing iron from 1 to 5 muM resulted in a marked increase in intracellular oxidative stress and increased GSH levels. Increased GSH levels were due to increased synthesis. Further increases in iron concentration led to significant reduction in both reduced (GSH) and total (GSH + (2 x GSSG)) glutathione. Cell exposure to high iron concentrations (20-80 muM) was associated with a marked decrease in the GSH/GSSG molar ratio and the GSH half-cell reduction potential. Moreover, increasing iron from 40 to 80 muM resulted in loss of cell viability. Iron loading did not change GSH reductase activity but induced significant increases in GSH peroxidase and GSH transferase activities. The changes in GSH homeostasis reported here recapitulate several of those observed in Parkinson's disease substantia nigra. These results support a model by which progressive iron accumulation leads to a progressive decrease in GSH content and cell reduction potential, which finally results in impaired cell integrity. (C) 2004 Elsevier Inc. All rights reserved.

Norris EH, Giasson BI, Lee VMY. 2004. Alpha-Synuclein: Normal Function and Role in Neurodegenerative DiseasesVolume 60. p 17-54 . Stem Cells in Development and Disease: Current Topics in Developmental Biology. Abstract: Synucleins are a family of small, highly charged proteins expressed predominantly in neurons. Since their discovery and characterization during the last decade, much has been learned about their structure, potential functions, interactions with other proteins, and roles in disease. One of these proteins, alpha-synuclein (alpha-syn), is the major building block of pathological inclusions that characterize many neurodegenerative disorders, including Parkinson's disease (PD), dementia with Lewy bodies (DLB), and neurodegeneration with brain iron accumulation type I (NBIA-1), which collectively are termed synucleinopathies. Furthermore, genetic and biological studies support a role for alpha-syn in the pathophysiology of these diseases. Therefore, research must be continued in order to better understand the functions of the synuclein proteins under normal physiological conditions as well as their role in diseases. (C) 2004 Elsevier Inc.

Normandin L, Beaupre LA, Salehi F, St-Pierre A, Kennedy G, Mergler D, Butterworth RE, Philippe S, Zayed J. 2004. Manganese distribution in the brain and neurobehavioral changes following inhalation exposure of rats to three chemical forms of manganese. Neurotoxicology 25(3):433-441. Abstract: The central nervous system is an important target for manganese (Mn) intoxication in humans; it may cause neurological symptoms similar to Parkinson's disease. Manganese compounds emitted from the tailpipe of vehicles using methylcyclopentadienyl manganese tricarbonyl (MMT) are primarily Mn phosphate, Mn sulfate, and Mn

phosphate/ sulfate mixture. The purpose of this study is to compare the patterns of Mn distribution in various brain regions (olfactory bulb, frontal parietal cortex, globus pallidus, striatum and cerebellum) and other tissues (lung, liver kidney, testis) and the neurobehavioral damage following inhalation exposure of rats to three Mn species. Rats (n = 15 rats per Mn species) were exposed 6 h per day, 5 days per week for 13 consecutive weeks to metallic Mn, Mn phosphate or Mn phosphate/ sulfate mixture at about 3000 mug m(-3) and compared to controls. At the end of the exposure period, spontaneous motor activity was measured for 36 h using a computerized autotrack system. Mn in tissues was determined by instrumental neutron activation analysis (INAA). The Mn concentrations in the brain were significantly higher in rats exposed to Mn phosphate and Mn phosphate/sulfate mixture than in control rats or rats exposed to metallic Mn. Exposure to Mn phosphate/sulfate mixture caused a decrease in the total ambulatory count related to locomotor activity. Our results confirm that Mn species and solubility have an influence on the brain distribution of Mn in rats. (C) 2003 Elsevier Inc. All rights reserved.

Noble M. 2004. The possible role of myelin destruction as a precipitating event in Alzheimer's disease. Neurobiol Aging 25(1):25-31.
Abstract: The number of hypotheses that have been put forward to explain the pathogenesis of Alzheimer's disease (AD) often seems beyond count. Many are sufficiently well-reasoned, and in some cases supported by some evidence, as to suggest that they are part of the picture of this important clinical problem (or collection of problems, as discussed later). How many of them will pave the way to important clinical interventions is, however, far from clear. One of the intriguing suggestions that has been made by multiple investigators is that an important component of AD is damage to myelin [4,5,13,18,29,36,43,50,55,66,87,98, 102,109]. Such a hypothesis is attractive for multiple reasons, as discussed in the review in this issue of Neurobiology of Aging by Bartzokis [3]. As discussed in this review, oligodendrocytes, the myelin-producing denizens of the CNS, are a strikingly vulnerable population of cells. In particular, susceptibility to death induced by oxidative stress, inflammatory cytokines and excitoxic neurotransmitters have all been extensively documented in this population. As loss of myelin compromises axonal impulse conduction, the neurological consequences of myelin destruction are considerable. The frequency with which destruction of myelin is seen in the CNS is so great, and occurs over such a wide range of disorders, as to likely qualify myelin disruption as the single largest category of clinically important CNS damage. Patients with AD do show increased latency of evoked potential responses [100], consistent with myelin-related abnormalities. The review by Bartzokis takes previous suggestions that loss of myelin may be an important component of understanding AD to the further step of suggesting that damage to oligodendrocytes may represent a critical initiating step in this important disease. He points out that the late-myelinating neocortical regions are the most vulnerable to the development of AD lesions, with the neurons most susceptible to neurodegeneration being the small diameter cortico-cortical axons that myelinate late in life [13,54,101,103]. Certainly a loss of oligodendrocytes might be expected to have profound effects not only on neuronal function, but also on neuronal viability. There are multiple indications that oligodendrocytes and/or the progenitor cells from which they are derived, also can provide trophic support for neurons (reviewed in [34]). Striatal oligodendrocyte-type-2 astrocyte progenitor cells (also known as oligodendrocyte precursor cells, and here abbreviated as O-2A/ OPCs) have been reported to enhance the survival of substantia nigra neurons through secreted factors [95,99]. O-2A/OPC lineage cells from the optic nerve can enhance retinal ganglion cell survival in vitro [70], basal forebrain oligodendrocytes enhance the survival of cholinergic neurons from this same brain region [22,24], and cortical O-2A/OPC lineage cells increase the in vitro survival of cortical neurons [112]. Expression of genes and proteins encoding multiple trophic factors has been observed in oligodendrocytes, including insulin-like growth factor-I, nerve growth factor, brain-derived neurotrophic factor, neurotrophin-3 and neurotrophin-4/5 [22,23,33]. Still other proteins that have been suggested

to be produced by oligodendrocytes include neuregulin-1 [16,30,83,108], GDNF [97], FGF-9 [72] and members of the TGF family [21,68]. In addition, the properties of oligodendrocytes, discussed in detail in the review of Bartzokis, as major contributors of CNS cholesterol and as the central storehouse of CNS iron also may be of great relevance to the development of AD pathology.

Nakatani N, Aburatani H, Nishimura K, Semba J, Yoshikawa T. 2004. Comprehensive expression analysis of a rat depression model. Pharmacogenomics Journal 4(2):114-126.
Abstract: Herein we report on a large-scale analysis of gene expression in the 'learned helplessness' (LH) rat model of human depression, using DNA microarrays. We compared gene expression in the frontal cortex (FC) and hippocampus (HPC) of untreated controls, and LH rats treated with saline (LH-S), imipramine or fluoxetine. A total of 34 and 48 transcripts were differentially expressed in the FC and HPC, respectively, between control and LH-S groups. Unexpectedly, only genes for NADH dehydrogenase and zinc transporter were altered in both the FC and HPC, suggesting limited overlap in the molecular processes from specific areas of the brain. Principal component analysis revealed that sets of upregulated metabolic enzyme genes in the FC and downregulated genes for signal transduction in the HPC can distinguish clearly between depressed and control animals, as well as explain the responsiveness to antidepressants. This comprehensive data could help to unravel the complex genetic predispositions involved in human depression.

Nakamichi N, Oikawa H, Kambe Y, Yoneda Y. 2004. Relevant modulation by ferrous ions of N-methyl-D-aspartate receptors in ischemic brain injuries. Current Neurovascular Research 1(5):429-440.
Abstract: Activation of the N-inethyl-D-aspartate (NMDA) receptor would induce rapid opening of an ion channel permeable to Ca2+ ions across cell membranes, followed by an increase in the concentration of free Ca2+ ions in the cytoplasm and subsequent signaling cascade from the cytoplasin to the nucleus for consolidation of a transient extracellular signal carried by L-glutamate in the central nervous system. Both neuronal plasticity and cell death have been shown to involve intracellular free Ca2+ ions incorporated through this receptor-operated cation channel in the brain. On the other hand, iron is also abundant in the brain, with an essential role in mechanisms underlying maintenance of cellular integrity and function. Ferrous ions are believed to participate in neuronal cell death through generation of reactive oxygen species in ischemic brain injuries, for instance, while ferrous but not ferric ions are shown to block the influx of Ca2+ ions across NMDA receptor channels in cultured neurons. In this review article, we will summarize the possible relationship between iron and NMDA receptor channels in mechanisms associated with neuronal cell death in brains with ischemia.

Mwanjewe J, Grover AK. 2004. Role of transient receptor potential canonical 6 (TRPC6) in non-transferrin-bound iron uptake in neuronal phenotype PC12 cells. Biochem J 378:975-982.
Abstract: Cells take up transferrin-bound iron or NTBI (non-transferrin-bound iron). After treatment with NGF (nerve growth factor), PC12 cells exhibited a neuronal phenotype and an increase in the NTBI uptake (Fe-55 (2+) or Fe-55(3+)). We loaded the cells with the dye calcein, whose fluorescence increases in the presence of Ca2+ but is quenched with Fe2+ or Fe2+. When examined using calcein fluorescence or radioactive iron, DAG (diacylglycerol)-stimulated NTBI entry was more in NGF-treated PC12 cells compared with untreated cells. All experiments were performed at 1.5 mM extracellular Ca2+. Nramp2 (natural-resistance-associated macrophage protein 2) mRNA expression did not change after the NGF treatment. Expression of the bivalent cation entry protein TRPC6 (transient receptor potential canonical 6) was detected only in the NGF-trearted cells. To verify that increased NTBI uptake depended on TRPC6, we examined whether transfecting HEK-293 (human embryonic kidney 293) cells with TRPC6 also increased the NTBI (Fe-55) uptake. We also cotransfected

HEK-293 cells with two plasmids, one expressing TRPC6 and the other expressing the fluorescent protein DsRED2 to identify the transfected cells. Challenging the calcein-loaded HEK-293 cells (which intrinsically express the alpha(1)-adrenergic receptors) with phenylephrine or a cell-permeant DAG increased the fluorescence signal more rapidly in transfected cells compared with untransfected cells. However, when iron (Fe2+ and Fe2+) was added before adding phenylephrine or DAG, the fluorescence intensity decreased more rapidly in transfected cells compared with untransfected cells, thereby indicating a greater stimulation of the NTBI uptake in cells expressing TRPC6. We postulate that the increase in the NTBI entry into neuronal PC12 cells is through TRPC6, a pathway that is unique since it is receptor-stimulated. Since neuronal cells express TRPC6, this pathway may have a role in neurotoxicity.

Munishkina LA, Cooper EM, Uversky VN, Fink AL. 2004. The effect of macromolecular crowding on protein aggregation and amyloid fibril formation. J Mol Recognit 17(5):456-464.
Abstract: Macromolecular crowding is expected to have several significant effects on protein aggregation; the major effects will be those due to excluded volume and increased viscosity. In this report we summarize data demonstrating that macromolecular crowding may lead to a dramatic acceleration in the rate of protein aggregation and formation of amyloid fibrils, using the protein a-synuclein. The aggregation of a-synuclein has been implicated as a critical factor in development of Parkinson's disease. Various types of polymers, from neutral polyethylene glycols and polysaccharides (Ficolls, dextrans) to inert proteins, are shown to accelerate a-synuclein fibrillation. The stimulation of fibrillation increases with increasing length of polymer, as well as increasing polymer concentration. At lower polymer concentrations (typically up to similar to100mg/ml) the major effect is ascribed to excluded volume, whereas at higher polymer concentrations evidence of opposing viscosity effects become apparent. Pesticides and metals, which are linked to increased risk of Parkinson's disease by epidemiological studies, are shown to accelerate alpha-synuclein fibrillation under conditions of molecular crowding. Copyright (C) 2004 John Wiley Sons, Ltd.

Moos T, Morgan EH. 2004. The Metabolism of Neuronal Iron and Its Pathogenic Role in Neurological Disease - ReviewVolume 1012. p 14-26. Redox-Active Metals in Neurological Disorders: Annals of the New York Academy of Sciences.
Abstract: Neurons need iron, which is reflected in their expression of the transferrin receptor. The concurrent expression of the ferrous iron transporter, divalent metal transporter I (DMT1), in neurons suggests that the internalization of transferrin is followed by detachment of iron within recycling endosomes and transport into the cytosol via DMT1. To enable DMT1-mediated export of iron from the endosome to the cytosol, ferric it-on must be reduced to its ferrous form, which could be mediated by a ferric reductase. The presence of nontransferrin-bound iron in brain extracellular fluids suggests that neurons can also take up iron in a transferrin-free form. Neurons are thought to be devoid of ferritin in many brain regions in which there is an association between iron accumulation and cellular damage, for example, neurons of the substantia nigra pars compacta. The general lack of ferritin together with the prevailing expression of the transferrin receptor indicates that iron acquired by activity of transferrin receptors is directed toward immediate use in relevant metabolic processes, is exported, or is incorporated into complexes other than ferritin. Iron has long been considered to play a significant role in exacerbating degradation processes in brain tissue subjected to acute damage and neurodegenerative disorders. In brain ischemia, the damaging role of iron may depend on the inhibition of detoxifying enzymes responsible for catalyzing the oxidation of ferrous iron. Brain ischemia may also lead to an increase in iron supply to neurons as transferrin receptor expression by brain capillary endothelial cells is increased. Pharmacological blockage of the transferrin receptor/DMT1-mediated uptake could be a target to prevent further it-on uptake. In

chronic neurodegenerative settings, a deleterious role of iron is suggested since cases of Alzheimer's disease, Parkinson's disease, and Huntington's disease have a significantly higher accumulation of iron in affected regions. Dopaminergic neurons are rich in neuromelanin, shown to be more redox-active in Parkinson's disease cases. Iron-containing inflammatory cells may, however, account for the main portion of iron present in neurodegenerative disorders. More knowledge about iron metabolism in normal and diseased neurons is warranted as this may identify pharmaceutical targets to improve neuronal iron management.

Moore SA, Huckerby TN, Gibson GL, Fullwood NJ, Turnbull S, Tabner BJ, El-Agnaf OMA, Allsop D. 2004. Both the D-(+) and L-(-) enantiomers of nicotine inhibit A beta aggregation and cytotoxicity. Biochemistry (Mosc) 43(3): 819-826.
Abstract: The underlying cause of Alzheimer's disease is thought to be the aggregation of monomeric beta-amyloid (Abeta), through a series of toxic oligomers, which forms the mature amyloid fibrils that accumulate at the center of senile plaques. It has been reported that L-(-)-nicotine prevents Abeta aggregation and toxicity, and inhibits senile plaque formation. Previous NMR studies have suggested that this could be due to the specific binding Of L-(-)-nicotine to histidine residues (His(6), His(13), and His(14)) in the peptide. Here, we have looked at the effects of both of the L-(-) and D-(+) optical enantiomers of nicotine on the aggregation and cytotoxicity of Abeta(1-40). Surprisingly, both enantiomers inhibited aggregation of the peptide and reduced the toxic effects of the peptide on cells. In NMR studies with Abeta(1-40), both enantiomers of nicotine were seen to interact with the three histidine residues. Overall, our data indicate that nicotine can delay A fibril formation and maintain a population of less toxic Abeta species. This effect cannot be due to a highly specific binding interaction between nicotine and Abeta, as previously thought, but could be due instead to weaker, relatively nonspecific binding, or to the antioxidant or metal chelating properties of nicotine. D-(+)-Nicotine, being biologically much less active than L-(-)-nicotine, might be a useful therapeutic agent.

Montplaisir J. 2004. Abnormal motor behavior during sleep. Sleep Medicine 5:S31-S34.
Abstract: Abnormal motor behaviors during sleep can be classified into four categories, ranging from myoclonic jerks to complex and integrated motor behaviors. There have been recent developments in several of these conditions, in particular restless legs syndrome (RLS) and rapid-eye-movement sleep behavior disorder (RBD). RLS is one of the major causes of insomnia. Familial aggregation of RLS has been demonstrated by several groups, and molecular genetics studies have suggested the presence of susceptibility genes on chromosomes 12q and 14q. Pharmacologic and brain imaging studies suggest the involvement of dopaminergic mechanisms in RLS, but recent work has focused on brain iron metabolism. Studies indicate that RBD patients may eventually develop Parkinson's disease (PD). Conversely, RBD has been found in patients already diagnosed with PD. Single-photon emission computed tomography and positron emission tomography studies have shown a decrease in binding to presynaptic dopamine transporter in both idiopathic RBD and PD. Patients with RBD (associated or unassociated with PD) also have neuropsychological deficits. RBD may therefore represent the prodrome of a neurodegenerative disease leading to multiple system atrophy and Lewy body dementia. Understanding the underlying pathophysiology of abnormal sleep motor behaviors may prove useful in the management of insomnia. (C) 2004 Published by Elsevier B.V. All rights reserved.

Mohamed GG, Zayed MA, El-Dien FAN, El-Nahas RG. 2004. IR, UV-Vis, magnetic and thermal characterization of chelates of some catecholamines and 4-aminoantipyrine with Fe(II) and Cu(II). Spectrochim Acta A Mol Biomol Spectrosc 60(8-9): 1775-1781.
Abstract: The dopamine derivatives participate in the regulation of wide variety of physiological functions in the human body and in medication life.

Increase and/or decrease in the concentration of dopamine in human body reflect an indication for diseases such as Schizophrenia and/or Parkinson diseases. alpha-Methyldopa (alpha-MD) in tablets is used in medication of hypertension. The Fe(III) and Cu(II) chelates with coupled products of adrenaline hydrogen tartarate (AHT), levodopa (LD), alpha-MD and carbidopa (CD) with 4-aminoantipyrine (4-AAP) are prepared and characterized. Different physico-chemical methods like IR, magnetic and UV-Vis spectra are used to investigate the structure of these chelates. Fe (III) form 1:2 (M:catecholamines) chelates while Cu(II) form 1:1 chelates. Catecholamines behave as a bidentate mono- or dibasic ligands in binding to the metal ions. IR spectra show that the catecholamines are coordinated to the metal ions in a bidentate manner with O,O donor sites of the phenolic -OH. Magnetic moment measurements reveal the presence of Fe (III) chelates in octahedral geometry while the Cu(II) chelates are square planar. The thermal decomposition of Fe(III) and Cu(II) complexes is studied using thermogravimetric (TGA) and differential thermal analysis (DTA) techniques. The water molecules are removed in the first step followed immediately by decomposition of the ligand molecules. The activation thermodynamic parameters, such as, energy of activation, enthalpy, entropy and free energy change of the complexes are evaluated and the relative thermal stability of the complexes are discussed. (C) 2003 Elsevier B.V. All rights reserved.

Mirecki A, Fitzmaurice P, Ang L, Kalasinsky KS, Peretti FJ, Aiken SS, Wickham DJ, Sherwin A, Nobrega JN, Forman HJ, Kish SJ. 2004. Brain antioxidant systems in human methamphetamine users. J Neurochem 89(6): 1396-1408.
Abstract: Animal data suggest that the widely abused psychostimulant methamphetamine can damage brain dopamine neurones by causing dopamine-dependent oxidative stress; however, the relevance to human methamphetamine users is unclear. We measured levels of key antioxidant defences [reduced (GSH) and oxidized (GSSG) glutathione, six major GSH system enzymes, copper-zinc superoxide dismutase (CuZnSOD), uric acid] that are often altered after exposure to oxidative stress, in autopsied brain of human methamphetamine users and matched controls. Changes in the total (n = 20) methamphetamine group were limited to the dopamine-rich caudate (the striatal subdivision with the most severe dopamine loss) in which only activity of CuZnSOD (+ 14%) and GSSG levels (+ 58%) were changed. In the six methamphetamine users with severe (- 72 to - 97%) caudate dopamine loss, caudate CuZnSOD activity (+ 20%) and uric acid levels (+ 63%) were increased with a trend for decreased (- 35%) GSH concentration. Our data suggest that brain levels of many antioxidant systems are preserved in methamphetamine users and that GSH depletion, commonly observed during severe oxidative stress, might occur only with severe dopamine loss. Increased CuZnSOD and uric acid might reflect compensatory responses to oxidative stress. Future studies are necessary to establish whether these changes are associated with oxidative brain damage in human methamphetamine users.

Mazzio EA, Reams RR, Soliman KFA. 2004. The role of oxidative stress, impaired glycolysis and mitochondrial respiratory redox failure in the cytotoxic effects of 6-hydroxydopamine in vitro. Brain Res 1004(1-2):29-44.
Abstract: The neurotoxin, 6-hydroxydopamine (6-OHDA) has been implicated in the neurodegenerative process of Parkinson's disease. The current study was designed to elucidate the toxicological effects of 6-OHDA on energy metabolism in neuroblastoma (N-2A) cells. The toxicity of 6-OHDA corresponds to the total collapse of anaerobic/aerobic cell function, unlike other mitochondrial toxins such as MPP+ that target specific loss of aerobic metabolism. The toxicity of 6-OHDA paralleled the loss of mitochondrial oxygen (02) consumption (MOC), glycolytic activity, ATP, H+ ion gradients, membrane potential and accumulation of the autoxidative product, hydrogen peroxide (H2O2). Removing H2O2 with nonenzymatic stoichiometric scavengers, such as carboxylic acids, glutathione and catalase yielded partial protection. The rapid removal of H2O2 with pyruvate or catalase restored only anaerobic glycolysis, but did not

reverse the loss of MOC, indicating mitochondrial impairment is independent of H2O2. The H2O2 generated by 6-OHDA contributed toward the loss of anaerobic glycolysis through lipid peroxidation and lactic acid dehydrogenase inhibition. The ability of 6-OHDA to maintain oxidized cytochrome c (CYT-C-OX) in its reduced form (CYT-C-RED), appears to play a role in mitochondrial impairment. The reduction of CYT-C by 6-OHDA, was extensive, occurred within minutes, preceded formation of H2O2 and was unaffected by catalase or superoxide dismutase. At similar concentrations, 6-OHDA readily altered the valence state of iron [Fe(Ill)] to Fe(H), which would also theoretically sustain CYT-C in its reduced form. In isolated mitochondria, 6-OHDA had negligible effects on complex 1, inhibited complex 11 and interfered with complex III by maintaining the substrate, CYT-C in a reduced state. 6-OHDA caused a transient and potent surge in isolated cytochrome oxidase (complex IV) activity, with rapid recovery as a result of 6-OHDA recycling CYT-C-OX to CYT-C-RED. Typical mitochondrial toxins such as MPP+, azide and antimycin appeared to inhibit the catalytic activity of ETC enzymes. In contrast, 6-OHDA alters the redox of the cytochromes, resulting in loss of substrate availability and obstruction of oxidation-reduction events. Complete cytoprotection against 6-OHDA toxicity and restored MOC was achieved by combining catalase with CYT-C (horse heart). In summary, CYT-C reducing properties are unique to catecholamine neurotransmitters, and may play a significant role in selective vulnerability of dopaminergic neurons to mitochondrial insults. (C) 2004 Elsevier B.V. All rights reserved.

Mattson MP. 2004. Metal-Catalyzed Disruption of Membrane Protein and Lipid Signaling in the Pathogenesis of Neurodegenerative DisordersVolume 1012. p 37-50. Redox-Active Metals in Neurological Disorders: Annals of the New York Academy of Sciences.
Abstract: Membrane lipid peroxidation and oxidative modification of various membrane and associated proteins (e.g., receptors, ion transporters and channels, and signal transduction and cytoskeletal proteins) occur in a range of neurodegenerative disorders. This membrane-associated oxidative stress (MAOS) is promoted by redox-active metals, most notably iron and copper. The mechanisms whereby different genetic and environmental factors initiate MAOS in specific neurological disorders are being elucidated. In Alzheimer's disease (AD), the amyloid beta-peptide generates reactive oxygen species and induces MAOS, resulting in disruption of cellular calcium homeostasis. In Parkinson's disease (PD), mitochondrial toxins and perturbed ubiquitin-dependent proteolysis may impair ATP production and increase oxyradical production and MAOS. The inheritance of polyglutamine-expanded huntingtin may promote neuronal degeneration in Huntington's disease (HD), in part, by increasing MAOS. Increased MAOS occurs in amyotrophic lateral sclerosis (ALS) as the result of genetic abnormalities (e.g., Cu/Zn-superoxide dismutase mutations) or exposure to environmental toxins. Levels of iron are increased in vulnerable neuronal populations in AD and PD, and dietary and pharmacological manipulations of iron and copper modify the course of the disease in mouse models of AD and PD in ways that suggest a role for these metals in disease pathogenesis. An increasing number of pharmacological and dietary interventions are being identified that can suppress MAOS and neuronal damage and improve functional outcome in animal models of AD, PD, HD, and ALS. Novel preventative and therapeutic approaches for neurodegenerative disorders are emerging from basic research on the molecular and cellular actions of metals and MAOS in neural cells.

Martin RCG, Hughes K, Doll MA, Lan Q, Martini BS, Lissowska J, Rothman N, Hein DW. 2004. Method for determination of (-102C > T) single nucleotide polymorphism in the human manganese superoxide dismutase promoter. Bmc Genetics 5.
Abstract: Background: Manganese superoxide dismutase (MnSOD) plays a critical role in the detoxification of mitochondrial reactive oxygen species constituting a major cellular defense mechanism against agents that induce oxidative stress. The MnSOD promoter contains an activator protein-2

(AP-2) binding site that modifies transcription of MnSOD. Mutations have been identified in the proximal region of the promoter in human tumor cell lines. One of these mutations (- 102C> T) has been shown to change the binding pattern of AP-2 leading to a reduction in transcriptional activity. The aim of our study was to develop a method to identify and determine the frequency of this (102C> T) polymorphism in human tissues. Results: A new TaqMan allelic discrimination genotype method was successfully applied to genomic DNA samples derived from blood, buccal swabs, snap frozen tissue and paraffin blocks. The polymorphism was shown to be in Hardy-Weinberg Equilibrium in an evaluation of 130 Caucasians from Warsaw, Poland: 44 (33.8%) were heterozygous and 6 (4.6%) were homozygous for - 102T. Conclusion: This report represents the first description of the MnSOD - 102C> T polymorphism in human subjects by a novel Taqman allelic discrimination assay. This method should enable molecular epidemiological studies to evaluate possible associations of this polymorphism with malignancies and other diseases related to reactive oxygen species.

Martin FL, Williamson SJM, Paleologou KE, Allsop D, El-Agnaf OMA. 2004. alpha-synuclein and the pathogenesis of Parkinson's disease. Protein and Peptide Letters 11(3):229-237.
Abstract: Lesions known as Lewy bodies (LBs) and Lewy neurites (LNs) characterise brains of Parkinson's disease (PD) patients. Intracellular aggregation of alpha-synuclein (alpha-syn) appears to play a key role in the generation of LBs and LNs. Such aggregation in the presence of redox metals may initiate Fenton reaction-mediated generation of reactive oxygen species (ROS). ROS thus generated may result in cytotoxic mechanisms such as the induction of DNA single-strand breaks.

Marciani P, Trotti D, Hediger MA, Monticelli G. 2004. Modulation of DMT1 activity by redox compounds. J Membr Biol 197(2):91-99.
Abstract: Iron(II) exacerbates the effects of oxidative stress via the Fenton reaction. A number of human diseases are associated with iron accumulation including ischemia-reperfusion injury, inflammation and certain neurodegenerative diseases. The functional properties and localization in plasma membrane of cells and endosomes suggest an important role for the divalent metal transporter DMT1 (also known as DCT1 and Nramp2) in iron transport and cellular iron homeostasis. Although iron metabolism is strictly controlled and the activity of DMT1 is central in controlling iron homeostasis, no regulatory mechanisms for DMTI have been so far identified. Our studies show that the activity of DMT1 is modulated by compounds that affect its redox status. We also show that both iron and zinc are transported by DMTI when expressed in Xenopus laevis oocytes. Radiotracer uptake and electrophysiological measurements revealed that H2O2 and Hg2+ treatments result in substantial inhibition of DMT1. These findings may have a profound relevance from a physiological and pathophysiological standpoint.

Mandel S, Youdim MBH. 2004. Catechin polyphenols: Neurodegeneration and neuroprotection in neurodegenerative diseases. Free Radic Biol Med 37(3): 304-317.
Abstract: Neurodegeneration in Parkinson's, Alzheimer's, and other neurodegenerative diseases seems to be multifactorial, in that a complex set of toxic reactions including inflammation, glutamatergic neurotoxicity, increases in iron and nitric oxide, depletion of endogenous antioxidants, reduced expression of trophic factors, dysfunction of the ubiquitin-proteasome system, and expression of proapoptotic proteins leads to the demise of neurons. Thus, the fundamental objective in neurodegeneration and neuroprotection research is to determine which of these factors constitutes the primary event, the sequence in which these events occur, and whether they act in concurrence in the pathogenic process, This has led to the current notion that drugs directed against a single target will be ineffective and rather a single drug or cocktail of drugs with pluripharmacological properties may be more suitable. Green tea catechin polyphenols, formerly thought to be simple radical scavengers, are now

considered to invoke a spectrum of cellular mechanisms of action related to their neuroprotective activity. These include pharmacological activities like iron chelation, scavenging of radicals, activation of survival genes and cell signaling pathways, and regulation of mitochondrial function and possibly of the ubiquitin-proteasome system. As a consequence these compounds are receiving significant attention as therapeutic cytoprotective agents for the treatment of neurodegenerative and other diseases. (C) 2004 Elsevier Inc. All rights reserved.

Mandel S, Weinreb O, Amit T, Youdim MBH. 2004. Cell signaling pathways in the neuroprotective actions of the green tea polyphenol (-)-epigallocatechin-3-gallate: implications for neurodegenerative diseases. J Neurochem 88(6): 1555-1569.
Abstract: Accumulating evidence supports the hypothesis that brain iron misregulation and oxidative stress (OS), resulting in reactive oxygen species (ROS) generation from H2O2 and inflammatory processes, trigger a cascade of events leading to apoptotic/necrotic cell death in neurodegenerative disorders, such as Parkinson's (PD), Alzheimer's (AD) and Huntington's diseases, and amyotrophic lateral sclerosis (ALS). Thus, novel therapeutic approaches aimed at neutralization of OS-induced neurotoxicity, support the application of ROS scavengers, transition metals (e.g. iron and copper) chelators and non-vitamin natural antioxidant polyphenols, in monotherapy, or as part of antioxidant cocktail formulation for these diseases. Both experimental and epidemiological evidence demonstrate that flavonoid polyphenols, particularly from green tea and blueberries, improve age-related cognitive decline and are neuroprotective in models of PD, AD and cerebral ischemia/reperfusion injuries. However, recent studies indicate that the radical scavenger property of green tea polyphenols is unlikely to be the sole explanation for their neuroprotective capacity and in fact, a wide spectrum of cellular signaling events may well account for their biological actions. In this article, the currently established mechanisms involved in the beneficial health action and emerging studies concerning the putative novel molecular neuroprotective activity of green tea and its major polyphenol (-)-epigallocatechin-3-gallate (EGCG), will be reviewed and discussed.

Mandel S, Maor G, Youdim MBH. 2004. Iron and alpha-synuclein in the substantia nigra of MPTP-treated mice: effect of neuroprotective drugs R-apomorphine and green tea polyphenol (-)-epigallocatechin-3-gallate. J Mol Neurosci 24(3):401-416.
Abstract: One of the prominent pathological features of Parkinson's disease (PD) is the abnormal accumulation of iron in the substantia nigra pars compacta (SNpc), in the reactive microglia, and in association with neuromelanin, within the melanin-containing dopamine (DA) neurons. Lewy body, the morphological hallmark of PD, is composed of lipids, redox-active iron, and aggregated alpha-synuclein, concentrating in its peripheral halo and ubiquitinated, hyperphosphorylated, neurofilament proteins. The capacity of free iron to enhance and promote the generation of toxic reactive oxygen radicals has been discussed numerous times. Recent observations, that iron induces aggregation of inert alpha-synuclein to toxic aggregates, have reinforced the critical role of iron in oxidative stress-induced pathogenesis of DA neuron degeneration and protein degradation via ubiquitination. N-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP)- and 6-hydroxydopamine-induced neurodegeneration in rodents and nonhuman primates is associated with increased presence of iron and alpha-synuclein in the SNpc. The accumulation of iron in MPTP-induced neurodegeneration has been linked to nitric oxide-dependent mechanism, resulting in degradation of prominent iron regulatory proteins by ubiquitination. Radical scavengers such as R-apomorphine and green tea catechin polyphenol (-)-epigallocatechin-3-gallate, as well as the recently developed brain-permeable VK-28 series derivative iron chelators, which are neuroprotective against these neurotoxins in mice and rats, prevent the accumulation of iron and alpha-synuclein in SNpc. This study supports the notion that a combination of iron chelation and antioxidant therapy, as emphasized on several occasions, might be a significant approach to

neuroprotection in PD and other neurodegenerative diseases.

Lowe R, Pountney DL, Jensen PH, Gai WP, Voelcker NH. 2004. Calcium(II) selectively induces alpha-synuclein annular oligomers via interaction with the C-terminal domain. Protein Sci 13(12):3245-3252.
Abstract: alpha-Synuclein filaments are the major component of intracytoplasmic inclusion bodies characteristic of Parkinson's disease and related disorders. The process of alpha-synuclein filament formation proceeds via intermediate or protofibrillar species, each of which may be cytotoxic. Because high levels of calcium(II) and other metal ions may play a role in disease pathogenesis, we investigated the influence of calcium and other metals on alpha-synuclein speciation. Here we report that calcium(II) and cobalt(II) selectively induce the rapid formation of discrete annular alpha-synuclein oligomeric species. We used atomic force microscopy to monitor the aggregation state of alpha-synuclein after 1 d at 4degreesC in the presence of a range of metal ions compared with the filament formation pathway in the absence of metal ions. Three classes of effect were observed with different groups of metal ions: (1) Copper(II), iron(III), and nickel(II) yielded 0.8-4 nm spherical particles, similar to alpha-synuclein incubated without metal ions; (2) magnesium(II), cadmium (II), and zinc(II) gave larger, 5-8 nm spherical oligomers; and, (3) cobalt (II) and calcium(II) gave frequent annular oligomers, 70-90 nm in diameter with calcium(II) and 22-30 nm in diameter with cobalt(II). In the absence of metal ions, annular oligomers ranging 45-90 nm in diameter were observed after 10 d incubation, short branched structures appeared after a further 3 wk and extended filaments after 2-3 mo. Previous studies have shown that a-synuclein calcium binding is mediated by the acidic C terminus. We found that truncated alpha-synuclein (1-125), lacking the C-terminal 15 amino acids, did not form annular oligomers upon calcium addition, indicating the involvement of the calcium-binding domain.

Love R. 2004. Dietary iron in Parkinson's disease: a double edged sword. Lancet Neurology 3(12):699.

Louis ED, Applegate LM, Factor-Litvak P, Parides MK, Andrews L. 2004. Essential tremor - Occupational exposures to manganese and organic solvents. Neurology 63(11):2162-2164.
Abstract: Occupational exposures to manganese and organic solvents cause parkinsonism as well as prominent action tremor, resembling essential tremor (ET), yet their association with ET has not been studied. These chemicals cause cerebellar pathology. Cerebellar changes have been linked with ET. Using lifetime occupational histories, the authors demonstrated that occupational exposures were similar in cases and controls, which does not support an etiologic link between occupational exposures to these chemicals and ET.

Lorenzl S, Calingasan N, Yang LC, Albers DS, Shugama S, Gregorio J, Krell HW, Chirichigno J, Joh T, Beal MF. 2004. Matrix metalloproteinase-9 is elevated in 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine-induced Parkinsonism in mice. Neuromolecular Medicine 5(2):119-131.
Abstract: Matrix metalloproteinases (MMPs) are proteolytic enzymes capable of degrading components of the extracellular matrix. Recent evidence has implicated MMPs in the pathogenesis of neurodegenerative diseases as Alzheimer's disease and amyotrophic lateral sclerosis. In this study, we investigated the involvement of MMP-9 (gelatinase B) in the 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) mouse model of Parkinson's disease using zymography, immunohistochemistry, and Western blot analysis. The activity of MMP-9 was upregulated at 3 h after MPTP injection in the striatum and after 24 h in the substantia nigra. Although MMP-9 expression decreased in the striatum by 72 h, it remained elevated in the substantia nigra compared to controls up to 7 d after MPTP administration. Immunohistochemistry showed that neurons and microglia are the source of MMP-9 expression after MPTP administration to mice. Treatment with a hydroxamate-based MMP inhibitor, Ro 28-2653 significantly reduced dopamine depletion and loss of tyrosine hydroxylase

immunoreactive neurons in the substantia nigra pars compacta. MMP-9 expression as measured via zymography in the substantia nigra was reduced by the MMP inhibitor. These results indicate that MMP-9 is induced after MPTP application in mice and that pharmacologic inhibition of MMPs protects against MPTP neurotoxicity.

Lo HS, Chiang HC, Lin AMY, Chiang HY, Chu YC, Kao LS. 2004. Synergistic effects of dopamine and Zn2+ on the induction of PC12 cell death and dopamine depletion in the striatum: possible implication in the pathogenesis of Parkinson's disease. Neurobiol Dis 17(1):54-61.
Abstract: The mechanism that underlies the progressive degeneration of the dopaminergic neurons in Parkinson's disease (PD) is not clear. The Zn2 + level in the substantia nigra of Parkinson's patients is increased. However, it is unknown whether Zn2+ has a role in the degeneration of dopaminergic neurons. This study identifies an interaction between dopamine and Zn2+ that induces cell death. When PC12 cells were pretreated with Zn2+ before dopamine treatment, dopamine and Zn2+ synergistically increased cell death, while Zn2+ and H2O2 had only additive effects on cell death. The synergistic effect appeared to be caused by increased apoptosis rather than necrosis. The synergistic effect was specific for Zn2+. The synergistic effect was inhibited by thiol antioxidants but was not significantly affected by calcium channel blockers. There is a similar synergistic effect when dopamine and Zn2+ were coinfused into the striatum, resulting in striatal dopamine content depletion in vivo. Thus, both dopamine oxidation and Zn2+ are possibly linked to the degeneration of dopaminergic neurons. (C) 2004 Elsevier Inc. All rights reserved.

Liang LP, Patel M. 2004. Iron-sulfur enzyme mediated mitochondrial superoxide toxicity in experimental Parkinson's disease. J Neurochem 90(5): 1076-1084.
Abstract: Mitochondrial oxidative stress is thought to be an important pathological mediator of neuronal death in Parkinson's disease. However, the precise mechanism by which mitochondrial oxidative stress mediates the death of dopaminergic neurons of the substantia nigra remains unclear. We tested the idea that neuronal damage in the MPTP (1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine) model of Parkinson's disease results, in part, from superoxide radical toxicity via inactivation of an iron-sulfur (Fe-S) protein, mitochondrial aconitase. Administration of MPTP in mice resulted in inactivation of mitochondrial aconitase, but not fumarase in the substantia nigra. MPTP treatment mobilized an early mitochondrial pool of iron detectable by bleomycin chelation that coincided with mitochondrial aconitase inactivation. MPTP-induced mitochondrial aconitase inactivation, iron accumulation and dopamine depletion were significantly attenuated in transgenic mice overexpressing mitochondrial Sod2 and exacerbated in partial deficient Sod2 mice. These results suggest that mitochondrial aconitase may be an important early source of mitochondrial iron accumulation in experimental Parkinson's disease, and that superoxide radical toxicity manifested by oxidative inactivation of mitochondrial aconitase may play a pathogenic role in Parkinson's disease.

Li SG, Crooks PA, Wei SC, De Leon J. 2004. Toxicity of dipyridyl compounds and related compounds. Crit Rev Toxicol 34(5):447-460.
Abstract: Five dipyridyl isomers, 2,2'-, 2,3-, 2,4'-, 3,3'-, and 4,4'-dipyridyl, are products resulting from the pyrolytic degradation of tobacco products and degradation of the herbicide paraquat, and therefore may be present in the environment. In this article, the toxicological properties of these dipyridyl isomers in humans and animals are reviewed. Epidemiological studies suggest that cancerous skin lesions in workers involved in the manufacturing of paraquat may be associated with exposure to dipyridyl compounds. Experimental animal studies suggest that dipyridyl isomers may have several toxicological effects. Three of the dipyridyl isomers (the 2,2', 2,4', and 4,4' isomers) appear to be inducers of some metabolic enzymes. The 2,2'-dipyridyl isomer, an iron chelator, appears to influence vasospasm in primate models of stroke. The cytotoxic effects of 2,2-dipyridyl on several leukemia cell lines have been reported, and a potent

teratogenic effect of 2,2'-dipyridyl has been observed in rats. Based on the results of paraquat studies in experimental animal models, it has been proposed that paraquat may have deleterious effects on dopaminergic neurons. These findings support the epidemiological evidence that paraquat exposure may be associated with the development of Parkinson's disease. Studies designed to determine an association between paraquat exposure and Parkinson's disease are complicated by the possibility that metabolic changes may influence the neurotoxicity of paraquat and/or its metabolites. Preliminary unpublished data in mice show that 300-mg/kg doses of 2,2'-dipyridyl are neurotoxic, and 300-mg/kg doses of 2,4'- and 4,4'-dipyridyls are lethal. These results are consistent with earlier studies in Sherman rats using high 2,2'- and 4,4'-dipyridyl doses. New studies are needed to further explore the toxicological properties of dipyridyls and their potential public health impact.

Li GJJ, Zhang LL, Lu L, Wu P, Zheng W. 2004. Occupational exposure to welding fume among welders: Alterations of manganese, iron, zinc, copper, and lead in body fluids and the oxidative stress status. J Occup Environ Med 46 (3):241-248.
Abstract: Welders in this study were selected from a vehicle manufacturer; control subjects were from a nearby food factory. Airborne manganese levels in the breathing zones of welders and controls were 1.45 +/- SD1.08 mg/m(3) and 0.11 +/- 0.07 mug/m(3), respectively. Serum levels of manganese and iron in welders were 4.3-fold and 1.9-fold, respectively, higher than those of controls. Blood lead concentrations in welders increased 2.5-fold, whereas serum zinc levels decreased 1.2-fold, in comparison with controls. Linear regression revealed the lack of associations between blood levels of five metals and welder's age. Furthermore, welders had erythrocytic superoxide dismutase activity and serum malondialdehyde levels 24% less and 78% higher, respectively, than those of controls. These findings suggest that occupational exposure to welding fumes among welders disturbs the homeostasis of trace elements in systemic circulation and induces oxidative stress.

Levenson CW, Tassabehji NA. 2004. Iron and ageing: an introduction to iron regulatory mechanisms. Ageing Research Reviews 3(3):251-263.
Abstract: While there have been significant advances made in our understanding of the cellular and molecular mechanisms that regulate iron absorption, transport, storage, and utilization, the effect of ageing on these mechanisms and the role of iron in the ageing process is not fully understood. Thus, this review will provide an overview of the iron regulatory mechanisms that may be a factor in the ageing process. Additional reviews in this volume represent an attempt to explore the very latest information on the regulation of iron with a particular emphasis on age-related pathology including mitochondrial function, Parkinson's disease, Alzheimer's disease, stroke, and cardiovascular disease. (C) 2004 Elsevier Ireland Ltd. All rights reserved.

Levenson CW, Cutler RG, Ladenheim B, Cadet JL, Hare J, Mattson MP. 2004. Role of dietary iron restriction in a mouse model of Parkinson's disease. Exp Neurol 190(2):506-514.
Abstract: There is a growing body of evidence suggesting that iron chelation may be a useful therapy in the treatment of Parkinson's Disease (PD). Experiments were designed to test the impact of dietary iron availability on the pathogenic process and functional outcome in a mouse model of PD. Mice were fed diets containing low (4 ppm) or adequate (48 ppm) amounts of iron for 6 weeks before the administration of MPTP, a mitochondrial toxin that damages nigrostriatal dopaminergic neurons and induces Parkinson-like symptoms. Low dietary iron increased serum total iron binding capacity ($P<0.001$). Consistent with neuronal protection, iron restriction increased sphingomyelin C16:0 and decreased ceramide C16:0. However, there was a 35% decrease in striatal dopamine (DA) in iron-restricted mice. Motor behavior was also impaired in these animals. In vitro studies suggested that severe iron restriction could lead to p53-mediated neuronal apoptosis. Administration of MPTP reduced striatal DA

(P<0.01) and impaired motor behavior in iron-adequate mice. However, in iron-restricted mice, striatal dopamine levels and motor behavior were unchanged compared to saline-treated mice. Thus, while reduced iron may provide protection against PD-inducing insults such as MPTP, the role of iron in the synthesis of DA and neuronal survival should be considered, particularly in the development of iron-chelating agents to be used chronically in the clinical setting. (C) 2004 Elsevier Inc. All rights reserved.

Lee EN, Cho HJ, Lee CH, Lee D, Chung KC, Paik SR. 2004. Phthalocyanine tetrasulfonates affect the amyloid formation and cytotoxicity of alpha-synuclein. Biochemistry (Mosc) 43(12):3704-3715.
Abstract: alpha-Synuclein is a pathological component of Parkinson's disease by constituting the filamentous component of Lewy bodies. Phthalocyanine (Pc) effects on the amyloidosis of alpha-synuclein have been examined. The copper complex of phthalocyanine tetrasulfonate (PcTS-Cu2+) caused the self-oligomerization of alpha-synuclein while Pc-Cu2+ did not affect the protein, indicating that introduction of the sulfonate groups was critical for the selective protein interaction. The PcTS-Cu2+ interaction with alpha-synuclein has occurred predominantly at the N-terminal region of the protein with a K-d of 0.83 muM apart from the hydrophobic NAC (non-Abeta component of Alzheimer's disease amyloid) segment. Phthalocyanine tetrasulfonate (PcTS) lacking the intercalated copper ion also showed a considerable affinity toward alpha-synuclein with a K-d of 3.12 muM, and its binding site, on the other hand, was located at the acidic C-terminus. These mutually exclusive interactions between PcTS and PcTS-Cu2+ toward alpha-synuclein resulted in distinctive features on the kinetics of protein aggregation, morphologies of the final aggregates, and their in vitro cytotoxicities. The PcTS actually suppressed the fibrous amyloid formation of alpha-synuclein, but it produced the chopped-wood-looking protein aggregates. The aggregates showed rather low toxicity (9.5%) on human neuroblastoma cells (SH-SY5Y). In fact, the PcTS was shown to effectively rescue the cell death of alpha-synuclein overexpressing cells caused by the lactacystin treatment as a proteasome inhibitor. The anti-aggregative and anti-amyloidogenic properties of PcTS were also demonstrated with alcohol dehydrogenase, glutathione S-transferase, and amyloid beta/A4 protein under their aggregative conditions. The PcTS-Cu2+, on the other hand, promoted the protein aggregation of alpha-synuclein, which gave rise to the fibrillar protein aggregates whose cytotoxicity became significant to 35.8%. Taken together, the data provided in this study indicate that PcTS/PcTS-Cu2+ could be considered as possible candidates for the development of therapeutic or prophylactic strategies against the alpha-synuclein-related neurodegenerative disorders.

Kurosaki R, Muramatsu Y, Kato H, Araki T. 2004. Biochemical, behavioral and immunohistochemical alterations in MPTP-treated mouse model of Parkinson's disease. Pharmacol Biochem Behav 78(1):143-153.
Abstract: The biochemical, behavioral and immunohistochemical manifestations were investigated in mice subjected to four experimental schedules with 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) hydrochloride treatment. The mice were treated intraperitoneally with MPTP (20 mg/kg in saline) four times a day at 2-h intervals showed severe and persistent depletions of dopamine, 3,4-dihydroxyphenylacetic acid (DOPAC) and homovanillic acid (HVA) in the striatum and behavioral deficits, as compared with those (1) treated with MPTP (15 mg/kg in saline ip) once a day for 14 consecutive days; (2) MPTP (30 mg/kg in saline ip) twice a day for five consecutive days; and (3) MPTP (10 mg/ kg in saline ip) four times a day at 1-h intervals for two consecutive days. The immunohistochemical study has shown that the acute treatment with MPTP caused severe loss of tyrosine hydroxylase (TH)- and dopamine transporter (DAT)-immunoreactive dopaminergic neurons and marked increase in glial fibrillary acidic protein (GFAP)-immunoreactive astrocytes in the striatum and the substantia nigra. Thus acute treatment of mice with MPTP was accompanied by sustained nigral degeneration and motor abnormalities. Furthermore, our results with Cu/ Zn-superoxide dismutase

(Cu/Zn-SOD) and manganese superoxide dismutase (Mn-SOD) immunostainings suggest that altered capacity of free radicals quenching may play a key role in the development of the neurons and interneuron damage after MPTP neurotoxicity. Thus, our findings provide valuable information on age-related disease progression and mechanisms of neurodegeneration. (C) 2004 Elsevier Inc. All rights reserved.

Kotamraju S, Tampo Y, Kalivendi SV, Joseph J, Chitambar CR, Kalyanaraman B. 2004. Nitric oxide mitigates peroxide-induced iron-signaling, oxidative damage, and apoptosis in endothelial cells: role of proteasomal function? Arch Biochem Biophys 423(1):74-80.
Abstract: In this mini-review, oxidant-induced transferrin receptor-mediated iron-signaling and apoptosis are described in endothelial and neuronal cells exposed to a variety of oxidative stresses. The role of nitric oxide and nitration in the regulation of iron homeostasis and oxidant-induced apoptosis is described. The interrelationship between oxidative stress, iron-signaling, and nitric oxide-dependent proteasomal function provides a rational mechanism that connects both oxidative and nitrative modifications. (C) 2004 Elsevier Inc. All rights reserved.

Koller WC, Lyons KE, Truly W. 2004. Effect of levodopa treatment for parkinsonism in welders - A double-blind study. Neurology 62(5):730-733.
Abstract: Background: Manganese is known to cause a parkinsonian syndrome similar clinically to Parkinson disease (PD). L-Dopa responsiveness is a hallmark of PD; however, L-dopa's effect on manganese-induced parkinsonism is not well defined. Objective: To access the efficacy and safety of L-dopa therapy in a double-blind, randomized, placebo-controlled trial. Methods: Thirteen patients with manganese-induced parkinsonism were evaluated in a cross-over study with a modified Unified PD Rating Scale (UPDRS), timed walk test, tapping, and global clinical impression scores. Adverse reactions were assessed. Results: There was no significant difference between placebo and L-dopa for any measure: motor UPDRS, 27.4 vs 28.8; walk time, 16.6 seconds vs 17.7 seconds; tapping right hand, 69.5 vs 64.7; and tapping left hand, 66.8 vs 64.4. There were no differences in the global impression scores. Adverse reactions occurred similarly in the two groups, including headaches, drowsiness, and diarrhea. Conclusions: L-Dopa therapy is not effective for the management of parkinsonism in welders. L-dopa unresponsiveness may be useful to distinguish manganese-induced parkinsonism from Parkinson disease.

Kitazawa M, Anantharam V, Kanthasamy A, Kanthasamy AG. 2004. Dieldrin promotes proteolytic cleavage of poly(ADP-Ribose) polymerase and apoptosis in dopaminergic cells: Protective effect of mitochondrial anti-apoptotic protein Bcl-2. Neurotoxicology 25(4):589-598.
Abstract: Previously, we demonstrated that the organochlorine pesticide dieldrin induces mitochondrial depolarization, caspase-3 activation and apoptosis in dopaminergic PC12 cells. We also demonstrated that protein kinase Cdelta (PKCdelta), a member of a novel PKC family of proteins, is proteolytically activated by caspase-3 to mediate apoptotic cell death processes. In the present study, we have further characterized the protective effect of the major mitochondrial anti-apoptotic protein Bcl-2 against dieldrin-induced apoptotic events in dopaminergic cells. Exposure to dieldrin (30-100 muM) produced significant cytotoxicity and caspase-3 activation within 3 h in vector-transfected PC12 cells, whereas human Bcl-2-transfected PC12 cells were almost completely resistant to dieldrin-induced cytotoxicity and caspase-3 activation. Also, dieldrin (30-300 muM) treatment induced proteolytic cleavage of poly(ADP-ribose) polymerase (PARP), which was blocked by pretreatment with caspase-3 inhibitors Z-DEVD-FMK and Z-VAD-FMK. Additionally, dieldrin-induced chromatin condensation and DNA fragmentation were completely blocked in Bcl-2-overexpressed PC12 cells as compared to vector control cells. Together, these results clearly indicate that overexpression of mitochondrial anti-apoptotic protein protects against dieldrin-induced apoptotic cell death and further suggest that dieldrin primarily alters mitochondrial function to

initiate apoptotic cell death in dopaminergic cells. (C) 2003 Elsevier Inc. All rights reserved.

King JE, Benson FE, Zhang H, Allsop D. 2004. Alpha-synuclein protein implicated in Parkinson's disease binds copper in vitro. Neurobiol Aging 25:S182.

Kim YH. 2004. High signal intensities on T1-weighted MRI as a biomarker of exposure to manganese. Ind Health 42(2):111-115.
Abstract: Increased signal in T1-weighted images was observed in the experimental manganese (Mn) poisoning of the non-human primate and a patient with Mn neurointoxication. However, our study showed that the increased signals in magnetic resonance images (MRI) were highly prevalent (41.6%) in Mn-exposed workers. Especially 73.5% of the welders showed increased signal intensities. Blood Mn concentration correlated with pallidal index. These changes in MRI tend to disappear following the withdrawal from the source of Mn accumulation, despite permanent neurological damage. Thus increased signal intensities on a T1-weighted image reflect exposure to Mn, but not necessarily manganism. Our study also showed that the concentration of Mn required to produce increased signal intensities on MRI is much lower than the threshold necessary to result in overt clinical signs of manganism. Increased signal intensities in the globus pallidus were determined by Mn accumulation in the animal experiment. All these results strongly suggest that signal intensities in T1-weighted MRI reflect a target site dose in a biologically-based dose-response model. At which increase of signal intensity, the progression of manganism from Mn exposure occurs, however, remains to be solved.

Kim NH, Jeong MS, Choi SY, Kang JH. 2004. Oxidative modification of neurofilament-L by the Cu,Zn-superoxide dismutase and hydrogen peroxide system. Biochimie 86(8):553-559.
Abstract: Neurofilament-L (NF-L) is a major element of neuronal cytoskeletons and known to be important for their survival in vivo. Since oxidative stress might play a critical role in the pathogenesis of neurodegenerative diseases, we investigated the role of Cu,Zn-superoxide dismutase (SOD) in the modification of NF-L. When disassembled NF-L was incubated with Cu,Zn-SOD and H2O2, the aggregation of protein was proportional to the concentration of hydrogen peroxide. Cu,Zn-SOD/H2O2-mediated modification of NF-L was significantly inhibited by radical scavenger, spin trap agents and copper chelators. Dityrosine crosslink formation was obtained in Cu,Zn-SOD/H2O2-mediated NF-L aggregates. Antioxidant molecules, carnosine and anserine significantly inhibited the aggregation of NF-L and the formation of dityrosine. This study suggests that copper-mediated NF-L modification may be closely related to oxidative reactions which play a critical role in neurodegenerative diseases. (C) 2004 Elsevier SAS. All rights reserved.

Khan A, Ashcroft AE, Korchazhkina OV, Exley C. 2004. Metal-mediated formation of fibrillar ABri amyloid. J Inorg Biochem 98(12):2006-2010.
Abstract: British amyloid (ABri) peptide is precipitated as amyloid fibrils in pathological lesions which are characteristic of familial British dementia. Unlike for other amyloidogenic peptides which have been implicated in neurodegenerative disease, for example, Abeta in Alzheimer's disease and alpha synuclein in Parkinson's disease, nothing is yet known as to whether metals mediate the formation of ABri amyloid fibrils. We show herein that a concentration of ABri, which had not previously been shown to spontaneously form amyloid, formed fibrils when incubated for 12 months at 37 degreesC. The additional presence of Al(III), in particular, or Fe(III) increased significantly both the number and the size of the fibrillar amyloid deposits which were very similar in appearance to amyloid described in hippocampal plaques in familial British dementia. Co-incubation of ABri with either Zn(II) or Cu(II) precipitated the peptide but did not result in the formation of amyloid fibrils. (C) 2004 Elsevier Inc. All rights reserved.

Kaur D, Andersen J. 2004. Does cellular iron dysregulation play a causative role in Parkinson's disease? Ageing Research Reviews 3(3):327-343.

Abstract: Selective dopaminergic cell loss in Parkinson's disease is correlated with increased levels of cellular iron. It is still hotly debated as to whether the increase in iron is an upstream event which acts to promote neurodegeneration via formation of oxidative stress or whether iron accumulates as a by-product of the neuronal cell loss. Here we review evidence for loss of iron homeostasis as a causative factor in disease-associated neurodegeneration and the primary players which may be involved. A series of recent studies suggest that iron regulatory proteins (IRPs) coordinate both cellular iron levels and energy metabolism, both of which are disrupted in Parkinson's disease (PD) and may in turn contribute to increased levels of oxidative stress associated with the disease. Iron has also been recently been implicated in promotion of alpha-synuclein aggregation either directly or via increasing levels of oxidative stress suggesting an important role for it in Lewy body formation, another important hallmark of the disease. (C) 2004 Elsevier Ireland Ltd. All rights reserved.

Kang JH. 2004. Modification of Cu,Zn-superoxide dismutase by oxidized catecholamines. Journal of Biochemistry and Molecular Biology 37(3): 325-329.
Abstract: Oxidation of catecholamines may contribute to the pathogenesis of Parkinson's disease (PD). The effect of the oxidized products of catecholamines on the modification of Cu,Zn-superoxide dismutase (SOD) was investigated. When Cu,Zn-SOD was incubated with the oxidized 3,4-dihydroxyphenylalanine (DOPA) or dopamine, the protein was induced to be aggregated. The deoxyribose assay showed that hydroxyl radicals were generated during the oxidation of catecholamines in the presence of copper ion. Radical scavengers, azide, N-acetylcysteine, and catalase inhibited the oxidized catecholamine-mediated Cu,Zn-SOD aggregation. Therefore, the results indicate that free radicals may play a role in the aggregation of Cu,Zn-SOD. When Cu,Zn-SOD that had been exposed to catecholamines was subsequently analyzed by an amino acid analysis, the glycine and histidine residues were particularly sensitive. These results suggest that the modification of Cu,Zn-SOD by oxidized catecholamines might induce the perturbation of cellular antioxidant systems and led to a deleterious cell condition.

Kang JH. 2004. Modification and inactivation of human ceruloplasmin by oxidized DOPA. Bulletin of the Korean Chemical Society 25(5):625-628.
Abstract: Ceruloplasmin (CP), the blue oxidase present in all vertebrates, is the major copper-containing protein of plasma. It has been proposed that oxidation of L-3,4-dihydroxyphenylalanine (DOPA) may contribute to the pathogenesis of neurodegenerative disorders. The effect of the oxidized products of DOPA on the modification of human CP was investigated. When CP was incubated with the oxidized L-DOPA, the protein was induced to be aggregated and ferroxidase activity was decreased in a time-dependent manner. Radical scavengers and catalase significantly inhibited the oxidized DOPA-mediated CP aggregation. Copper chelatrors, Diethylenetriaminepenta acetic acid (DTPA) and Diethyldithiocarbamic acid (DDC), also inhibited the oxidative modification of CP. The results suggested that DOPA oxidation led to the formation of free radical and induced the CP aggregation.

Kalivendi SV, Cunningham S, Kotamraju S, Joseph J, Hillard CJ, Kalyanaraman B. 2004. alpha-synuclein up-regulation and aggregation during MPP+-induced apoptosis in neuroblastoma cells - Intermediacy of transferrin receptor iron and hydrogen peroxide. J Biol Chem 279(15):15240-15247.
Abstract: 1-Methyl-4-phenylpyridinium (MPP+) is a neurotoxin that causes Parkinson's disease in experimental animals and humans. Despite the fact that intracellular iron was shown to be crucial for MPP+-induced apoptotic cell death, the molecular mechanisms for the iron requirement remain unclear. We investigated the role of transferrin receptor (TfR) and iron in modulating the expression of alpha-synuclein (alpha-syn) in MPP+-induced oxidative stress and apoptosis. Results show that MPP+ inhibits mitochondrial complex-1 and aconitase activities leading to enhanced H2O2

generation, TfR expression and alpha-syn expression/aggregation. Pretreatment with cell-permeable iron chelators, TfR antibody (that inhibits TfR-mediated iron uptake), or transfection with glutathione peroxidase (GPx1) enzyme inhibits intracellular oxidant generation, alpha-syn expression/aggregation, and apoptotic signaling as measured by caspase-3 activation. Cells overexpressing alpha-syn exacerbated MPP+ toxicity, whereas antisense alpha-syn treatment totally abrogated MPP+-induced apoptosis in neuroblastoma cells without affecting oxidant generation. The increased cytotoxic effects of alpha-syn in MPP+-treated cells were attributed to inhibition of mitogen-activated protein kinase and proteasomal function. We conclude that MPP+-induced iron signaling is responsible for intracellular oxidant generation, alpha-syn expression, proteasomal dysfunction, and apoptosis. Relevance to Parkinson's disease is discussed.

Johnson MA, Kuo YM, Westaway SK, Parker SM, Ching KHL, Gitschier J, Hayflick SJ. 2004. Mitochondrial Localization of Human Pank2 and Hypotheses of Secondary Iron Accumulation in Pantothenate Kinase-Associated NeurodegenerationVolume 1012. p 282-298. Redox-Active Metals in Neurological Disorders: Annals of the New York Academy of Sciences.
Abstract: Mutations in the pantothenate kinase 2 gene (PANK2) lead to pantothenate kinase-associated neurodegeneration (PKAN, formerly Hallervorden-Spatz syndrome). This neurodegenerative disorder is characterized by iron accumulation in the basal ganglia. Pantothenate kinase is the first enzyme in the biosynthesis of coenzyme A from pantothenate (vitamin B-5). PANK2, one of four human pantothenate kinase genes, is uniquely predicted to be targeted to mitochondria. We demonstrate mitochondrial localization of PANK2 and speculate on mechanisms of secondary iron accumulation in PKAN. Furthermore, PANK2 uses an unconventional translational start codon, CUG, which is polymorphic in the general population. The variant sequence, CAG (allele frequency: 0.05), leads to skipping of the mitochondrial targeting signal and cytosolic localization of PANK2. This common variant may cause mitochondrial dysfunction and impart susceptibility to late-onset neurodegenerative disorders with brain iron accumulation, including Parkinson's disease.

Ji LN, Li HT, Luo XY, Zhang F, Hu HY, Hu J. 2004. The concentration of hydrogen peroxide generated during aggregation of alpha-synuclein in vitro is lower than 5 nmol/L. Chinese Journal of Chemistry 22(12):1440-1443.
Abstract: Using a fluorometric method with a detection limit of 5 nmol/L, here it is reported that albeit positive results were got from bovine serum albumin (BSA) and chicken ovalbun-fin (OVA) as published in literature, no detectable amount of hydrogen peroxide (H2O2) was generated during alpha-synuclein (alpha-Syn) aggregation in vitro even in the presence of transition metal ions Cu(II) or Fe(III). The results suggest that the concentration of H2O2 generated during aggregation of alpha-Syn in vitro be lower than 5 nmol/L beyond the detection limit of the adopted method and it is far too poor to be responsible for the cytotoxicity of alpha-Syn aggregates, thus allowing people to extensively elucidate the mechanism underlying neurotoxicities of the aggregates formed by some amyloidogenic proteins.

Jameson GNL, Jameson RF, Linert W. 2004. New insights into iron release from ferritin: direct observation of the neurotoxin 6-hydroxydopamine entering ferritin and reaching redox equilibrium with the iron core. Organic & Biomolecular Chemistry 2(16):2346-2351.
Abstract: Iron release from the iron storage protein ferritin has been studied extensively because of its important role in oxidative stress and its possible role in the progression of Parkinson's disease. For many years external indicators, notably strong iron(II) chelators, have been used to investigate this reaction. Such chelators can, however, drastically affect the electrochemical and thermodynamic properties of iron. The present study is unique in that it has been possible to follow a reaction taking place within the ferritin shell. This was made possible by our serendipitous discovery that, at physiological pHs, the oxidation product of 6-

hydroxydopamine (a deprotonated quinone) acts as its own indicator (G.N.L. Jameson and W. Linert, J. Chem. Soc., Perkin Trans. 2, 2001, 563-568). The redox equilibrium data and the kinetics of the formation of this red-coloured species can only be explained on the basis that reduction of the iron(III) takes place within the ferritin shell. This is, in fact, the first time that a reaction actually taking place inside the ferritin shell has been followed. It has also been established that, at least in vitro, all eight hydrophilic channels are capable of being simultaneously involved in the reaction. It has also been possible to calculate the rate of oxidation of the 6-hydroxydopamine within the ferritin and it is demonstrated that a redox equilibrium is established within the protein. Finally, evidence is provided confirming that chelators are in fact intrinsically linked to iron removal from ferritin.

Iova A, Garmashov A, Androuchtchenko N, Kehrer M, Berg D, Becker G, Garmashov Y. 2004. Postnatal decrease in substantia nigra echogenicity - Implications for the pathogenesis of Parkinson's disease. J Neurol 251(12): 1451-1454.
Abstract: Increased echogenicity of the substantia nigra (SN) on transcranial ultrasonography (TCS) is a typical sign in Parkinson's disease (PD). Detected in healthy adults it is assumed to represent a risk factor for nigral injury. We studied at which time point of brain maturation increased signal intensity may occur by performing TCS scans in 109 newborns and children aged 0-192 months. While newborns regularly exhibit SN hyperechogenicity, this echofeature decreases substantially during the first years of life. As SN echogenicity is related to the tissue iron content in adults our findings suggest a failure in SN iron metabolism in some children with increased echogenicity during development which can be disclosed by TCS.

Inoue T, Satoh E, Ohyashiki T. 2004. Analysis of mechanism of aluminum-induced cell injury - Caspase-3 is not a direct execution factor. Yakugaku Zasshi 124:257-260 .
Abstract: As is well known, that aluminum (Al) induces several neurodegenerative disorders including Alzheimer's disease, dialysis encephalopathy, amyotrophic lateral scleorosis and Parkinsonism-dementia of Guam, However, the mechanism by which Al induces cell death has not been clarified yet. In the present study, we demonstrated that Al(maltol)(3)-treatment of PC12 cells induces the cell death via apoptosis. A general caspase inhibitor zVAD-fmk effectively prevented this cell death, but a caspase-3 selective inhibitor zDEVD-fmk did not show the protective effect against the cell death. These results suggest that caspase-3 is not an executioner caspase in Al(maltol)(3)-induced cell death. This is further confirmed by the experimental results with serum-starved cells death. In addition, in the present study, we proposed the possibility that the reactive oxygen species(ROS) generated during the treatment play an important role for the onset of Al toxicity.

Ide-Ektessabi A, Kawakami T, Watt F. 2004. Distribution and chemical state analysis of iron in the Parkinsonian substantia nigra using synchrotron radiation micro beams. Nuclear Instruments & Methods in Physics Research Section B-Beam Interactions With Materials and Atoms 213:590-594.
Abstract: Metallic elements and their organic compounds have dynamic regulatory functions in cells. Iron concentrations have been observed in the neuromelanin granules in the substantia nigra of brain tissues of patients with Parkinson's disease. iron has been linked to cell death because of its potential to promote free radicals, leading to oxidative stress. In the present study, we have used synchrotron radiation X-ray fluorescence spectroscopy (SXRF) and Fe K-edge X-ray absorption near-edge structure (XANES) spectroscopy, to investigate distributions and,chemical states of iron. The samples were brain tissues from monkeys which had been injected with MPTP (1-metyl-4-phenyl-1,2,3,6-tetrahydropyridine). SXRF analyses were performed for elemental mapping, using 7.16 keV energy beam.. The chemical state analyses were performed between 7.16 and 7.12 keV energies. The lower limit was

chosen to be slightly above the Fe2+ absorption edge, in order to suppress the excitation of Fe3+. FeO (Fe2+) and Fe2O3 (Fe3+) powders were used for XANES analyses as reference samples. The data were measured in fluorescence mode for the biological specimens and in transmission mode for the reference samples. The results for the Fe2+/Fe3+ ratios from the neuromelanin granules showed significant variations, which were correlated with the level of iron concentration. Cells containing high level of iron had high level of Fe2+. With Fe2+ having been suggested to potentially promote more free radicals than Fe3+, the high concentrations of iron may be the critical factor leading to cell death due to the presence of more free radicals. (C) 2003 Elsevier B.V. All rights reserved.

Hung RJ, Boffetta P, Brennan P, Malaveille C, Gelatti U, Placidi D, Carta A, Hautefeuille A, Porru S. 2004. Genetic polymorphisms of MPO, COMT, MnSOD, NQO1, interactions with environmental exposures and bladder cancer risk. Carcinogenesis 25(6):973-978.
Abstract: Tobacco smoking and occupational exposure are major risk factors of bladder cancer via exposure to polycyclic aromatic hydrocarbons (PAHs) and aromatic amines, which lead to oxidative stress and DNA damage. Several enzymes, which play key roles in oxidative stress are polymorphic in humans. Myeloperoxidase (MPO) produces a strong oxidant for microbicidal activity, and activates carcinogens in tobacco smoke. Catechol-O-methyltransferase (COMT) catalyzes the methylation of endo- and xenobiotics and prevents redox cycling. NAD(P)H:quinone oxidoreductase (NQO1) catalyzes the two-electron reduction of quinoid compounds, which also protects cells from redox cycling. Manganese superoxide dismutase (MnSOD) protects cells from free radical injury. To test the hypothesis that the risk of bladder cancer can be influenced by polymorphisms in the genes that modulate oxidative stress, in particular by interacting with environmental carcinogens, we conducted a hospital-based case-control study among men in Brescia, Northern Italy. We recruited and interviewed 201 incident cases and 214 controls from 1997 to 2000. Occupational exposures to PAHs and aromatic amines were coded blindly by occupational physicians. Unconditional multivariate logistic regression was applied to model the association between genetic polymorphisms and bladder cancer risk and the effect of modifications of smoking and occupational exposures were evaluated. MPO G-463A homozygous variant was associated with a reduced risk of bladder cancer with an OR of 0.31 (95% CI = 0.12-0.80). MnSOD Val/Val genotype increased the risk of bladder cancer with OR of 1.91 (95% CI = 1.20-3.04), and there was a combined effect with smoking (OR = 7.20, 95% CI = 3.23-16.1) and PAH (OR = 3.02, 95% CI = 1.35-6.74). We did not observe an effect of COMT Val108Met polymorphism. These findings suggest that individual susceptibility of bladder cancer may be modulated by MPO and MnSOD polymorphisms, and that the combination of genetic factors involved in oxidative stress response with environmental carcinogens may play an important role in bladder carcinogenesis.

Huang XD, Moir RD, Tanzi RE, Bush AI, Rogers JT. 2004. Redox-Active Metals, Oxidative Stress, and Alzheimer's Disease PathologyVolume 1012. p 153-163. Redox-Active Metals in Neurological Disorders: Annals of the New York Academy of Sciences.
Abstract: Considerable evidence is mounting that dyshomeostasis of the redox-active biometals, Cu and Fe, and oxidative stress contribute to the neuropathology of Alzheimer's disease (AD). Present data suggest that metals can interact directly with Abeta peptide, the principal component of beta-amyloid that is one of the primary lesions in AD. The binding of metals to Abeta modulates several physiochemical properties of Abeta that are thought to be central to the pathogenicity of the peptide. First, We and others have shown that metals can promote the in vitro aggregation into tinctorial Abeta amyloid. Studies have confirmed that insoluble amyloid plaques in postmortem AD brain are abnormally enriched in Cu, Fe, and Zn. Conversely, metal chelators dissolve these proteinaceous deposits from postmortem AD brain tissue and attenuate cerebral Abeta amyloid burden in APP transgenic mouse models of AD. Second, we have

demonstrated that redox-active Cu(II) and, to a lesser extent, Fe(III) are reduced in the presence of Abeta with concomitant production of reactive oxygen species (ROS), hydrogen peroxide (H2O2) and hydroxyl radical (OH.). These Abeta/metal redox reactions, which are silenced by redox-inert Zn(II), but exacerbated by biological reducing agents, may lead directly to the widespread oxidation damages observed in AD brains. Moreover, studies have also shown that H2O2 mediates Abeta cellular toxicity and increases the production of both Abeta and amyloid precursor protein (APP). Third, the 5' untranslated region (5'UTR) of APP mRNA has a functional iron-response element (IRE), which is consistent with biochemical evidence that APP is a redox-active metalloprotein. Hence, the redox interactions between Abeta, APP, and metals may be at the heart of a pathological positive feedback system wherein Abeta amyloidosis and oxidative stress promote each other. The emergence of redox-active metals as key players in AD pathogenesis strongly argues that amyloid-specific metal-complexing agents and antioxidants be investigated as possible disease-modifying agents for treating this horrible disease.

Huang E, Ong WY, Connor JR. 2004. Distribution of divalent metal transporter-1 in the monkey basal ganglia. Neuroscience 128(3):487-496.
Abstract: An accumulation of iron occurs in the brain with age, and it is thought that this may contribute to the pathology of certain neurodegenerative diseases, including Parkinson's disease. In this study, we elucidated the distribution of divalent metal transporter-1 (DMT1) in the monkey basal ganglia by immunocytochemistry, and compared it with the distribution of ferrous iron in these nuclei by Turnbull's Blue histochemical staining. We observed a general correlation between levels of DMT1, and iron staining. Thus, regions such as the caudate nucleus, putamen, and substantia nigra pars reticulata contained dense staining of DMT1 in astrocytic processes, and were also observed to contain large numbers of ferrous iron granules. The exceptions were the globus pallidus externa and interna, which contained light DMT1 staining, but large numbers of ferrous iron granules. The thalamus, subthalamic nucleus, and substantia nigra pars compacta contained neurons that were lightly stained for DMT1, but few or no iron granules. The high levels of DMT1 expression in some of the nuclei of the basal ganglia, particularly the caudate nucleus, putamen, and substantia nigra pars reticulata, may account for the high levels of iron in these regions. (C) 2004 IBRO. Published by Elsevier Ltd. All rights reserved.

Hozumi I, Asanuma M, Yamada M, Uchida Y. 2004. Metallothioneins and neurodegenerative diseases. Journal of Health Science 50(4):323-331.
Abstract: A symposium on the clinical aspects of metallothioneins (MTs) in neurodegenerative diseases was held at the 2003 Society of Metallothionein Meeting in Gifu, Japan. The objectives of the symposium were to review and speculate on the potential roles of MTs, especially MT-3 in neurodegenerative diseases such as Alzheimer's disease (AD), Parkinson's disease (PD), amyotrophic lateral sclerosis (ALS) and spinocerebellar degeneration (SCD). Dr. Uchida discussed the controversial problem regarding the expression of MT-3 in AD brains. Dr. Asanuma addressed the function of MTs in the progression of PD and Dr. Hozumi described the therapeutic potential of MTs for ALS, while Dr. Yamada provided immunohistochemical findings of MT-3 in SCD for the first time. Although there are still controversial problems on MTs, this review provides proof that MTs are promising potential therapeutic targets for therapy for some neurodegenerative diseases.

Hokenson MJ, Uversky VN, Goers J, Yamin G, Munishkina LA, Fink AL. 2004. Role of individual methionines in the fibrillation of methionine-oxidized alpha-synuclein. Biochemistry (Mosc) 43(15):4621-4633.
Abstract: The aggregation of normally soluble alpha-synuclein in the dopaminergic neurons of the substantia nigra is a crucial step in the pathogenesis of Parkinson's disease. Oxidative stress is believed to be a contributing factor in this disorder. We have previously established that oxidation of all four methionine residues in alpha-synuclein (to the

sulfoxide, MetO) inhibits fibrillation of this protein in vitro and that the MetO protein also inhibits fibrillation of unmodified alpha-synuclein. Here we show that the degree of inhibition of fibrillation by MetO alpha-synuclein is proportional to the number of oxidized methionines. This was accomplished be selectively converting Met residues into Leu, prior to Met oxidation. The results showed that with one oxidized Met the kinetics of fibrillation were comparable to those for the control (nonoxidized), and with increasing numbers of methionine sulfoxides the kinetics of fibrillation became progressively slower. Electron microscope images showed that the fibril morphology was similar for all species examined, although fewer fibrils were observed with the oxidized forms. The presence of zinc was shown to overcome the Met oxidation-induced inhibition. Interestingly, substitution of Met by Leu led to increased propensity for aggregation (soluble oligomers) but slower formation of fibrils.

Hochstrasser H, Bauer P, Walter U, Behnke S, Spiegel J, Csoti I, Zeiler B, Bornemann A, Pahnke J, Becker G, Riess O, Berg D. 2004. Ceruloplasmin gene variations and substantia nigra hyperechogenicity in Parkinson disease. Neurology 63(10):1912-1917.
Abstract: Background: Transcranial ultrasound may be used to detect increased iron levels of the substantia nigra (SN) in patients with Parkinson disease (PD) and in control subjects. It is not known whether iron accumulation in PD is a primary or secondary phenomenon. However, sequence variations in genes involved in iron metabolism have been linked to basal ganglia disorders. One of these is ceruloplasmin (Cp), which is vitally involved in iron transport across the cell membrane. Methods: One hundred seventy-six patients with PD according to the UK Brain Bank criteria and 180 ethnically matched control subjects, who were previously examined for SN iron signal changes by transcranial ultrasound, were examined for mutations in the Cp gene using denaturing high-performance liquid chromatography and subsequent sequencing for verification of unequivocal signals. Immunohistochemistry of PD midbrains was performed to examine the presence of Cp in Lewy bodies. Results: Five novel missense variations were detected. One of these (I63T) was found in a single PD patient. A known variation (D554E) was significantly associated with PD and the ultrasound marker for increased SN iron levels. Moreover, a third sequence variation (R793H) was found to segregate with the ultrasound marker for increased iron levels in patients and control subjects. Immunohistochemistry demonstrated that Cp co-localizes with Lewy bodies in PD. Conclusions: Detection of sequence variations in a single Parkinson disease (PD) patient or associated with the ultrasound marker for increased substantia nigra iron levels and the presence of ceruloplasmin (Cp) immunoreactivity in Lewy bodies underline a suspected role for Cp in the pathogenesis of PD. Further functional analyses are warranted to investigate whether these variations are causally linked to the complex pathogenesis of PD in a subset of cases.

Hirata Y, Furuta K, Miyazaki S, Suzuki M, Kiuchi K. 2004. Anti-apoptotic and pro-apoptotic effect of NEPP11 on manganese-induced apoptosis and JNK pathway activation in PC12 cells. Brain Res 1021(2):241-247.
Abstract: Neurite outgrowth-promoting prostaglandins (NEPPs), cyclopentenone prostaglandin derivatives, are found to be neurotrophic. These small organic compounds promote neurite outgrowth of PC 12 cells and dorsal root ganglion explants in the presence of nerve growth factor, and prevent neuronal cell death of HT22 cells and cortical neurons induced by various stimuli. In this study, we examined whether NEPP11 prevents manganese-induced apoptosis of PC 12 cells. NEPP11 (5 muM) attenuated manganese-induced DNA fragmentation by approximately 50%. In addition, NEPP11 partially prevented manganese-induced c-Jun phosphorylation and c-Jun N-terminal kinase (JNK) phosphorylation determined by Western blotting. Inhibition of the JNK signaling pathway by NEPP11 appeared to be selective, because NEPP 11 did not inhibit manganese-induced activation of p38 mitogen-activated protein kinase (p38 MAPK), extracellular signal-regulated kinase1/2 (ERK1/2), MEK1/2 and p70 S6 kinase (p70S6K) in PC 12 cells. In contrast, NEPP11 alone was

toxic at higher concentrations (>10 muM) producing DNA fragmentation and activation of the JNK pathway. Molecular modifications of NEPP11 may strengthen its inhibitory effects on the JNK pathway while preventing its cytotoxicity, and thus may become a useful small molecule reagent for the treatment of manganese toxicity and other similar neurodegenerative processes. (C) 2004 Elsevier B.V. All rights reserved.

Higashi Y, Asanuma M, Miyazaki I, Hattori N, Mizuno Y, Ogawa N. 2004. Parkin attenuates manganese-induced dopaminergic cell death. J Neurochem 89 (6):1490-1497.
Abstract: Manganese as environmental factor is considered to cause parkinsonism and induce endoplasmic reticulum stress-mediated dopaminergic cell death. We examined the effects of manganese on parkin, identified as the gene responsible for familial Parkinson's disease, and the role of parkin in manganese-induced neuronal cell death. Manganese dose-dependently induced cell death of dopaminergic SH-SY5Y and CATH.a cells and cholinergic Neuro-2a cells, and that the former two cell types were more sensitive to manganese toxicity than Neuro-2a cells. Moreover, manganese increased the expression of endoplasmic reticulum stress-associated genes, including parkin, in SH-SY5Y cells and CATH.a cells, but not in Neuro-2a cells. Treatment with manganese resulted in accumulation of parkin protein in SH-SY5Y cells and its redistribution to the perinuclear region, especially aggregated Golgi complex, while in Neuro-2a cells neither expression nor redistribution of parkin was noted. Manganese showed no changes in proteasome activities in either cell. Transient transfection of parkin gene inhibited manganese- or manganese plus dopamine-induced cell death of SH-SY5Y cells, but not of Neuro-2a cells. Our results suggest that the attenuating effects of parkin against manganese- or manganese plus dopamine-induced cell death are dopaminergic cell-specific compensatory reactions associated with its accumulation and redistribution to perinuclear regions but not with proteasome system.

Hermida-Ameijeiras A, Mendez-Alvarez E, Sanchez-Iglesias S, Sanmartin-Suarez C, Soto-Otero R. 2004. Autoxidation and MAO-mediated metabolism of dopamine as a potential cause of oxidative stress: role of ferrous and ferric ions. Neurochem Int 45(1):103-116.
Abstract: The autoxidation and monoamine oxidase (MAO)-mediated metabolism of dopamine (3-hydroxytyramine; DA) cause a continuous production of hydroxyl radical ((OH)-O-.), which is further enhanced by the presence of iron (ferrous iron, Fe2+ and ferric ion, Fe3+). The accumulation of hydrogen peroxide (H2O2) in the presence of Fe2+ appears to discard the involvement of the Fenton reaction in this process. It has been found that the presence of DA significantly reduces the formation of thiobarbituric acid reagent substances (TBARS), which under physiological conditions takes place in mitochondrial preparations. The presence of DA is also able to reduce TBARS formation in mitochondrial preparations even in the presence of iron (Fe2+ and Fe3+). However, DA boosted the carbonyl content of mitochondrial proteins, which was further increased in the presence of iron (Fe2+ and Fe3+). This latter effect is also accompanied by a significant reduction in thiol content of mitochondrial proteins. It has also been observed how the pre-incubation of mitochondria with pargyline, an acetylenic MAO inhibitor, reduces the production of (OH)-O-. and increases the formation of TBARS. Although, the MAO-mediated metabolism of DA increases MAO-B activity, the presence of iron inhibits both MAO-A and MAO-B activities. Consequently, DA has been shown to be a double-edged sword, because it displays antioxidant properties in relation to both the Fenton reaction and lipid peroxidation and exhibits pro-oxidant properties by causing both generation (OH)-O-. and oxidation of mitochondrial proteins. Evidently, these pro-oxidant properties of DA help explain the long-term side effects derived from L-DOPA treatment of Parkinson's disease and its exacerbation by the concomitant use of DA metabolism inhibitors. (C) 2003 Elsevier Ltd. All rights reserved.

Heim C, Sontag TA, Kolasiewicz W, Ulrich F, Pardowitz I, Horn HJ, Gerlach M,

Riederer P, Sontag KH. 2004. Consequences of a single short lasting cerebral oligemia and the influence of iron injected into the substantia nigra or in the ventrolateral striatum of the rat. Trigger of Parkinson's disease pathogenesis? J Neural Transm 111(6):641-666.
Abstract: One BCCA-phase (bilateral clamping of carotid arteria) leads to an extensive release of striatal dopamine with a subsequent formation of free radicals (Heim et al., 200b). Early investigations did not show histological damage to cerebral structures after 24 and 60 min duration of a BCCA phase (Melzacka et al., 1994). The study here turned out that oligemic damage and an increase in iron (FeCl3) concentration in the ventral striatum was responsible for most of the defective performance of the animals investigated. Striatal damaged animals were unable to correct their deficient performance to the same extent as was possible for animals which had been damaged through BCCA and FeCl3 in the substantia nigra. Furthermore it turn out that with the use of a comprehensive behaviour profile which was able to gather 22 parameters simultaneously, 15 of these parameters did not correspond in the performance of the controls already after BCCA alone. Since during the ageing process, pathological effects may occur in vulnerable structures not only from disturbances to cerebral blood-perfusion but also from enrichment of iron in vulnerable structures (Connor, 1992) the question arose whether this situation did not reveal pathological mechanisms that might triggered the early symptoms of Parkinson's disease.

Heckl S, Pipkorn R, Nagele T, Vogel U, Kuker W, Voigt K. 2004. Molecular imaging: Bridging the gap between neuroradiollogy and neurohistology. Histol Histopathol 19(2):651-668.
Abstract: Historically, in vivo imaging methods have largely relied on imaging gross anatomy. More recently it has become possible to depict biological processes at the cellular and molecular level. These new research methods use magnetic resonance imaging (MRI), positron emission tomography (PET), near-infrared optical imaging, scintigraphy, and autoradiography in vivo and in vitro. Of primary interest is the development of methods using MRI and PET with which the progress of gene therapy in glioblastoma (herpes simplex virus-thymidine kinase) and Parkinson's disease can be monitored and graphically displayed. The distribution of serotonin receptors; in the human brain and the duration of serotonin- receptor antagonist binding can be assessed by PET. With PET, it is possible to localize neurofibrillary tangles (NFTs) and beta-amyloid senile plaques (APs) in the brains Of living Alzheimer disease (AD) patients. MR tracking of transplanted oligodendrocyte progenitors is feasible for determining the extent of remyelinization in myelin-deficient rats. Stroke therapy in adult rats with subventricular zone cells can be monitored by MRI. Transgene expression (B-galactosidase, tyrosinase, engineered transferrin receptor) can also be visualized using MRI. Macrophages can be marked with certain iron-containing contrast agents which, through accumulation at the margins of glioblastomas, ameliorate the visual demarcation in MRI. The use of near-infrared optical imaging techniques to visualize matrix-metalloproteinases and cathepsin B can improve the assessment of tumor aggressiveness and angiogenesis-inhibitory therapy. Apoptosis could be detected using near-infrared optical imaging representation of caspase 3 activity and annexin B. This review demonstrates the need for neurohistological research if further progress is to be made in the emerging but burgeoning field of molecular imaging.

Hasegawa T, Matsuzaki M, Takeda A, Kikuchi A, Akita H, Perry G, Smith MA, Itoyama Y. 2004. Accelerated alpha-synuclein aggregation after differentiation of SH-SY5Y neuroblastoma cells. Brain Res 1013(1):51-59.
Abstract: alpha-Synticlein (alpha-syn) is a major component of inclusion bodies in Parkinson's disease (PD) and other synucleinopathies. To clarify the possible roles of a-syn in the molecular pathogenesis of neurodegenerative diseases, we have established a novel cellular model based on the differentiation of SH-SY5Y cells that overexpress alpha-syn. In the presence of ferrous iron, differentiation of the cells led to the formation of large perinuclear inclusion bodies, which developed from

scattered small aggregates seen in undifferentiated cells. The iron-induced alpha-synpositive inclusions co-localized largely with ubiquitin, and some of them were positive for nitrotyrosine, lipid, gamma-tubulin and dynein. Notably, treatment with nocodazole, a microtubule depolymerizing agent, interrupted the aggregate formation but led to a concomitant increase of apoptotic cells. Therefore, it appears that an intracellular retrograde transport system via inicrotubules plays a crucial role in the aggregate formation and also that the aggregates may represent a cytoprotective response against noxious stimuli. This cellular model will enable better understanding of the molecular pathomechanisms of synucleinopathy. (C) 2004 Elsevier B.V. All rights reserved.

Hamai D, Bondy SC. 2004. Oxidative Basis of Manganese NeurotoxicityVolume 1012. p 129-141. Redox-Active Metals in Neurological Disorders: Annals of the New York Academy of Sciences.
Abstract: Exposure to excessive levels of manganese, an essential trace element, can evoke severe psychiatric and extrapyramidal motor dysfunction closely resembling Parkinson's disease. The clinical manifestations of manganese toxicity arise from focal injury to the basal ganglia. This region, characterized by intense consumption of oxygen and significant dopamine content, can incur mitochondrial dysfunction, depletion of levels of peroxidase and catalase, and catecholamine biochemical imbalances following manganese exposure. The site specificity of the pathology and the nature of the cellular damage caused by manganese have been attributed to its capacity to produce cytotoxic levels of free radicals. However, support for such a pro-oxidant role for manganese has been largely limited to inferences drawn from histopathological observations. More recently, research efforts into the molecular details of manganese toxicity have provided evidence of an etiological relationship between oxidative stress and manganese-related neurodegeneration. This review focuses on studies that evaluate the redox chemistry of manganese during the neurodegenerative process and its molecular consequences.

Gunter TE, Miller LM, Gavin CE, Eliseev R, Salter J, Buntinas L, Alexandrov A, Hammond S, Gunter KK. 2004. Determination of the oxidation states of manganese in brain, liver, and heart mitochondria. J Neurochem 88(2): 266-280.
Abstract: Excess brain manganese can produce toxicity with symptoms that resemble those of Parkinsonism and causes that remain elusive. Manganese accumulates in mitochondria, a major source of superoxide, which can oxidize Mn2+ to the powerful oxidizing agent Mn3+. Oxidation of important cell components by Mn3+ has been suggested as a cause of the toxic effects of manganese. Determining the oxidation states of intramitochondrial manganese could help to identify the dominant mechanism of manganese toxicity. Using X-ray absorbance near edge structure (XANES) spectroscopy, we have characterized the oxidation state of manganese in mitochondria isolated from brain, liver, and heart over concentrations ranging from physiological to pathological. Results showed that (i) spectra from different model manganese complexes of the same oxidation state were similar to each other and different from those of other oxidation states and that the position of the absorption edge increases with oxidation state; (ii) spectra from intramitochondrial manganese in isolated brain, heart and liver mitochondria were virtually identical; and (iii) under these conditions intramitochondrial manganese exists primarily as a combination of Mn2+ complexes. No evidence for Mn3+ was detected in samples containing more than endogenous manganese levels, even after incubation under conditions promoting reactive oxygen species (ROS) production. While the presence of Mn3+ complexes cannot be proven in the spectrum of endogenous mitochondrial manganese, the shape of this spectrum could suggest the presence of Mn3+ near the limit of detection, probably as MnSOD.

Grunblatt E, Mandel S, Jacob-Hirsch J, Zeligson S, Amariglo N, Rechavi G, Li J, Ravid R, Roggendorf W, Riederer P, Youdim MBH. 2004. Gene expression

profiling of parkinsonian substantia nigra pars compacta; alterations in ubiquitin-proteasome, heat shock protein, iron and oxidative stress regulated proteins, cell adhesion/cellular matrix and vesicle trafficking genes. J Neural Transm 111(12):1543-1573.
Abstract: Gene expression profiling of human substantia nigra pars compacta (SNpc) from Parkinson's disease (PD) patients, was examined employing high density microarrays. We identified alterations in the expression of 137 genes, with 68 down regulated and 69 up regulated. The down regulated genes belong to signal transduction, protein degradation (e.g. ubiquitin-proteasome subunits), dopaminergic transmission/ metabolism, ion transport, protein modification/phosphorylation and energy pathways/glycolysis functional classes. Up-regulated genes, clustered mainly in biological processes involving cell adhesion/ cytoskeleton, extracellular matrix components, cell cycle, protein modification/phosphorylation, protein metabolism, transcription and inflammation/stress (e.g. key iron and oxygen sensor EGLN1). One major finding in the present study is the particular decreased expression of SKP1A, a member of the SCF (E3) ligase complex specifically in the substantia nigra (SN) of sporadic parkinsonian patients, which may lead to a wide impairment in the function of an entire repertoire of proteins subjected to regulatory ubiquitination. These findings reveal novel players in the neurodegenerative scenario and provide potential targets for the development of novel drug compounds.

Gotz ME, Double K, Gerlach M, Youdim MBH, Riederer P. 2004. The Relevance of Iron in the Pathogenesis of Parkinson's DiseaseVolume 1012. p 193-208. Redox-Active Metals in Neurological Disorders: Annals of the New York Academy of Sciences.
Abstract: Investigations that revealed increased levels of iron in postmortem brains from patients with Parkinson's disease (PD) as compared to those from individuals not suffering from neurological disorders are reported. The chemical natures in which iron predominates in the brain and the relevance of neuromelanin for neuronal iron binding are discussed. Major findings have been that iron levels increase with the severity of neuropathological changes in PD, presumably due to increased transport through the blood-brain barrier in late stages of parkinsonism. Glial iron is mainly stored as ferric iron in ferritin, while neuronal iron is predominantly bound to neuromelanin. Iron overload may induce progressive degeneration of nigrostriatal neurons by facilitating the formation of reactive biological intermediates, including reactive oxygen species, and the formation of cytotoxic protein aggregates. There are indications that iron-mediated neuronal death in PD proceeds retrogradely. These results are also discussed with respect to their relevance for disease progression in relation to cytotoxic alpha-synuclein protofibril formation.

Gossuin Y, Muller RN, Gillis P. 2004. Relaxation induced by ferritin: a better understanding for an improved MRI iron quantification. NMR Biomed 17(7): 427-432.
Abstract: Ferritin, the iron storing protein, is known to darken T-2-weighted MRI. This darkening can be used to noninvasively measure iron content. However, ferritin's behavior is not the same in tissue as in solution, a discrepancy that remains unexplained by the recently developed theory matching the NMR properties of ferritin solutions. A better understanding of the relaxation induced by ferritin in tissue could help for the development of new MRI protocols of iron quantification. In this short review, the main relaxation properties of ferritin in solution and in tissue are presented together with a discussion of the possible reasons for the faster transverse relaxation observed in tissues. Copyright (C) 2004 John Wiley Sons, Ltd.

Gossuin Y, Burtea C, Monseux A, Toubeau G, Roch A, Muller RN, Gillis P. 2004. Ferritin-induced relaxation in tissues: An in vitro study. J Magn Reson Imaging 20(4):690-696.
Abstract: Purpose: To study in vitro the proton relaxation induced in tissues by ferritin, the iron-storing protein of mammals. Materials and Methods: Nuclear magnetic relaxation dispersion (NMRD) profiles of liver

and spleen from control and iron-overloaded mice are compared with NMRD profiles of ferritin and Fercayl(R)-a ferritin-like akaganeite particle-in aqueous solutions or in 1% agarose gel. Results: The relaxation of water protons induced by ferritin and Fercayl(R) in 1% agarose gel is comparable with the relaxation of aqueous solutions of the same compounds, but slower than the relaxation of liver and spleen. The gel is not a good model of tissues containing ferritin. The longitudinal NMRD profiles of control and iron-overloaded liver and spleen are almost identical: ferritin accumulation has only a slight effect on longitudinal relaxation. The transverse NMRD profiles of liver and spleen tissues are linear, but the slope of the linear regression is larger for iron-loaded organs than for control ones, which is a consequence of a higher ferritin concentration in the former. However, the correlation between the slope of the transverse NMRD profiles and the iron concentration is not very good, probably because transverse relaxation is modified by the clustering of ferritin in cells. Conclusion: It could be difficult to develop a general technique for the accurate quantification of ferritin-bound iron by nuclear magnetic resonance or magnetic resonance imaging.

Gorell JM, Peterson EL, Rybicki BA, Johnson CC. 2004. Multiple risk factors for Parkinson's disease. J Neurol Sci 217(2):169-174.
Abstract: Objective: To determine the relative contribution of various risk factors to the development of Parkinson's disease (PD). Methods: Ten variables that were independently associated with PD in a health system population-based case-control study of epidemiological risk factors for the disease were jointly assessed. Stepwise logistic regression, adjusted for sex, race and age was used to develop a multiple variate model that best predicted the presence of PD. The population attributable risk was estimated for each variable in the final model, as well as for all factors together. Results: The 10 initial variables included >20 years occupational exposure to manganese or to copper, individually; >20 years joint occupational exposure to either lead and copper, copper and iron, or lead and iron; a positive family history of PD in first- or second-degree relatives; occupational exposure to insecticides or herbicides; occupational exposure to farming; and smoking. Logistic regression resulted in a final model that included >20 years joint occupational exposure to lead and copper (p = 0.009; population attributable risk [PAR] = 3.9%), occupational exposure to insecticides (p = 0.002; PAR = 8.1%), a positive family history of PD in first- and second-degree relatives (p = 0.001; PAR = 12.4%), and smoking :5 30 pack-years or not smoking (p = 0.005; PAR = 41.4%). All four variables combined had a PAR = 54.1%. Conclusions: Our final model of PD risk suggests that occupational, environmental lifestyle and, likely, genetic factors, individually and collectively, play a significant role in the etiology of the disease. Clearly, additional risk factors remain to be determined through future research. (C) 2003 Elsevier B.V. All rights reserved.

Gorell JM, Coon S, Peterson EL, Gloi A, Pounds JG, Chettle DR. 2004. Total body lead burden and the risk of Parkinson's disease. Mov Disord 19:S270.

Gonzalez-Polo RA, Soler G, Rodriguezmartin A, Moran JM, Fuentes JM. 2004. Protection against MPP+ neurotoxicity in cerebellar granule cells by antioxidants. Cell Biol Int 28(5):373-380.
Abstract: The neuropathology associated with Parkinson's disease (PD) is thought to involve excessive production of free radicals, dopamine autoxidation, defects in glutathione peroxidase expression, attenuated levels of reduced glutathione, altered calcium homeostasis, excitotoxicity and genetic defects in mitochondrial complex I activity. While the neurotoxic mechanisms are vastly different for excitotoxins and 1-methyl-4-phenylpyridinium ion (MPP+), both are thought to involve free radical production, compromised mitochondrial activity and excessive lipid peroxidation. We show here that the levels of reactive oxygen species (ROS) and reactive nitrogen species (RNS) increased significantly after treatment of cultured cerebellar granule cells (CGCs) with 50 muM MPP+. Co-treatment with antioxidants such as ascorbate (ASC), catalase, alpha-tocopherol (alpha-TOH), coenzyme Q(10) (CoQ(10)) or superoxide

dismutase (SOD) rescued the cells from MPP+-induced death. MPP+-induced cell death was also abolished by co-treatment with nitric oxide synthase (NOS) inhibitors such as 7-nitroindazole (7-NI), 2-ethyl-2-thiopseudourea hydrobromide (EPTU) or S-methylisothiourea sulphate (MPTU). We also tested the protective effects of an iron chelator (deferoxamine mesylate, DFx) and a peroxynitrite scavenger (FcTTPS) and the results lend further support to the view that the free radical cytotoxicity plays an essential role in MPP+-induced death in primary cultures of CGC. (C) 2004 International Federation for Cell Biology. Published by Elsevier Ltd. All rights reserved.

Goldman SM, Quinlan PJ, Smith AR, Langston J, Tanner CM. 2004. Manganese exposure and risk of Parkinson's disease in twins. Mov Disord 19:S162.

Garcia-Borreguero D, Odin P, Schwarz C. 2004. Restless legs syndrome: an overview of the current understanding and management. Acta Neurol Scand 109(5):303-317.
Abstract: Over the last few years, major progress in research has improved our understanding of the restless legs syndrome (RLS). Although frequently under-diagnosed, several epidemiological studies have estimated its prevalence in western countries at 4-10% of the general population. Its diagnosis is usually made on a clinical basis, according to the criteria established by the International RLS Study Group (Mov Disord 1995; 10:634). Furthermore, major advances have also been achieved regarding our understanding of the pathophysiology of the disorder. Thus, several brain imaging studies, as well as pharmacological challenges, suggest the presence of a dopaminergic dysfunction playing a major role in its causation. In addition, a strong association has been discovered between brain iron deficiency and RLS. Eventually, dopaminergic drugs have shown therapeutic efficacy in various large-scale therapeutic trials, and, today, dopaminergic agonists represent the first line of treatment. In conclusion, these and other recent findings shed light on our understanding and management of one of the most common movement disorders.

Gandhe KR, Deshpande NR, Chaubal R. 2004. High accumulation of a heavy metal in Ravenelia esculenta: an edible rust. Asian Journal of Chemistry 16 (2):1197-1199.
Abstract: Ravenelia esculenta, an edible rust infecting Acacia eburnea is consumed with relish in Maharashtra. Metal analysis of the host and parasite showed high percentage of aluminium in the parasite namely Ravenelia esculenta. So more consumption of this rust may lead to metabolic disorders such as Alzheimer's disease, Parkinsonism etc. in human beings.

Ganadu ML, Aru M, Mura GM, Coi A, Mlynarz P, Kozlowski H. 2004. Effects of divalent metal ions on the alpha beta-crystallin chaperone-like activity: spectroscopic evidence for a complex between copper(II) and protein. J Inorg Biochem 98(6):1103-1109.
Abstract: alphaB-crystallin is a small heat shock protein, showing chaperone-like activity, that is expressed in the lens and in several other tissues. The role of some metal ions in the alphaB-crystallin biology starts to be well documented. In some neuro-degenerative pathologies, like Parkinson and Alzheimer's diseases, alphaB-crystallin is expressed at high levels. In the same pathologies an accumulation of divalent metal cations is observed. In order to investigate the interactions between human alphaB-crystallin and divalent metal ions, the effect of copper, zinc and calcium on the chaperone-like activity of the protein has been studied. Copper and zinc at concentrations 0.1 and 1 mM significantly increase the chaperone-like activity, whereas calcium I mM completely inhibits activity. Electron paramagnetic resonance (EPR) and circular dichroism (CD) spectra indicate the possible complex formation between Cu(II) and protein at physiological pH. Molecular modeling calculations, carried out for the probable Cu(II) binding site, suggest that a complex with three histidine residues is possible. (C) 2004 Elsevier Inc. All rights reserved.

Galazka-Friedman J, Bauminger ER, Koziorowski D, Friedman A. 2004. Mossbauer spectroscopy and ELISA studies reveal differences between. Parkinson's disease and control substantia nigra. Biochimica Et Biophysica Acta-Molecular Basis of Disease 1688(2):130-136.
Abstract: The possible role of iron in the degeneration of nervous cells in Parkinson's disease (PD) was studied with the use of Mossbauer spectroscopy (MS) and enzyme-linked immunoabsorbent assay (ELISA). Mossbauer data were obtained at 90 and 4.1 K from 21 samples of control and 9 samples of parkinsonian substantia nigra (SN). Mossbauer spectra were very similar to those observed in ferritin. Small differences were detected between the spectra obtained from PD and from control SN, and could be due to a slight difference in the composition of the ferritin-like iron cores or due to the presence of about 8% of non-ferritin-like iron in parkinsonian SN. ELISA studies from 11 controls and 6 parkinsonian SN showed a decrease in the concentration of L-chains in wet tissues of PD-SN compared to control SN. The decrease in the amount of L subunits may correspond to a decreased ability of this ferritin to keep iron in a safe form. Iron released from ferritin or neuromelanin (NM) may be the source of such iron, which may cause the difference in the Mossbauer spectra and may trigger oxidative stress leading to cell death. (C) 2003 Elsevier B.V. All rights reserved.

Friedman A, Galazka-Friedman J, Koziorowski D, Bauminger ER. 2004. Difference in nigral iron between Parkinson's disease and control is in the iron binding not in amount. Ann Neurol 56:S67.

Fraker PJ, Lill-Elghanian DA. 2004. The many roles of apoptosis in immunity as modified by aging and nutritional status. J Nutr Health Aging 8(1):56-63.
Abstract: Apoptosis plays a vital role in the elimination of anti-self clones, down regulation of immune responses and the killing of virally infected and malignant cells. There is ample evidence that as we age the immune system not only becomes less potent, but dysregulated which includes apoptotic dependent functions. Reductions in the production of naive T and B-cells, reduced cytolytic killing capacity, accumulation of larger numbers of malignant cells, enhanced inflammatory responses, etc., in the aged suggest that apoptosis is dysregulated. Changes in nutritional status can also alter apoptosis. A short period of zinc deficiency (ZD) in young adult mice greatly accelerated apoptosis among pre-B and pre-T cells by 50% to 300% providing a mechanistic explanation for the lymphopenia and thymic atrophy long associated with this and other nutritional deficiencies. Since apoptosis has been shown to be altered by aging and nutritional status, it seemed important to determine how ZD affected these processes in the aged mouse. It was quickly discovered that the pre-B cells were reduced by 80% in the 28 month aged mouse making further studies problematic. In marked contrast to suboptimal zinc, caloric restriction (CR) which when initiated in younger mice delayed the onset of autoimmunity and immunosenescence. CR appeared to also slow the aging of mitochondria and, thereby, reduced the release of reactive oxygen species that damage cells. Thus, it is probable that CR also helped maintain the integrity of mitochondria and apoptotic processes as mice aged. Though CR is not a very practical nutritional model for humans, the outcome of these studies reinforce the potential value of anti-oxidants in our diets. In contrast to their normal nutritional role some nutrients especially small amounts of free metals can induce apoptosis. There is considerable zinc in neurons. As will be discussed, a number of investigators think that this zinc is released during Alzheimer's, Parkinsons's, or brain injury and accelerates apoptosis in surrounding tissues causing greater damage. Data are discussed that indicate nanomoles of free zinc is, indeed, a potent inducer of apoptosis in a variety of tissues. In sum, there is no doubt that nutritional status as well as individual nutrients can modulate apoptosis and that their impact on cell death may become greater in the aged.

Fortunato G, Marciano E, Zarrilli F, Mazzaccara C, Intrieri M, Calcagno G, Vitale DF, La Manna P, Saulino C, Marcelli V, Sacchetti L. 2004. Paraoxonase and superoxide dismutase gene polymorphisms and noise-induced hearing

loss. Clin Chem 50(11):2012-2018.
Abstract: Background: Noise-induced cochlear epithelium damage can cause hearing loss in industrial workers. In experimental systems, noise induces the release of free radicals and may damage the cochlear sensorial epithelium. Therefore, genes involved in regulating the reactive oxygen species manganese-superoxide dismutase (SOD2) and the antioxidant paraoxonase (PON) could influence cochlea vulnerability to noise. We evaluated whether susceptibility to noise-induced hearing loss (NIHL) is associated with SOD2, PON1, and PON2 polymorphisms in workers exposed to prolonged loud noise. Methods: We enrolled 94 male workers from an aircraft factory in the study. The SOD2 gene Was screened by denaturing reversed-phase. HPLC, and the PON1 (Q192R and M55L) and PON2 (S311C) polymorphisms were analyzed by PCR amplification followed by digestion with restriction endonucleases. Results: Three known (A16V, IVS3-23T/G, and IVS3-60T/G) and two new SOD2 polymorphisms (IVS1+8A/G and IVS3+107T/A) were identified. Regression analysis showed that PON2 (SC+CC) [odds ratio (OR)=5.01; 95% confidence interval (CI), 1.11-22.54], SOD2 IVS3-23T/G and IVS3-60T/G (OR=5.09; 95% CI, 1.27-20.47), age (OR=1.22; 95% CI, 1.09-1.36), and smoking (OR = 49.49; 95% CI, 5.09-480.66) were associated with NIHL. No association was detected for PON1 (QQ+RR) and PON1 (LL) genotypes. Conclusions: Our data suggest that SOD2 and PON2 polymorphisms, by exerting variable local tissue antioxidant roles, could predispose to NIHL. However, caution should be exercised in interpreting these data given the small sample size and the difficulty in matching cases to controls regarding the overwhelming risk factor, i.e., smoking at least 10 cigarettes/day. (C) 2004 American Association for Clinical Chemistry.

Forte G, Bocca B, Senofonte O, Petrucci F, Brusa L, Stanzione P, Zannino S, Violante N, Alimonti A, Sancesario G. 2004. Trace and major elements in whole blood, serum, cerebrospinal fluid and urine of patients with Parkinson's disease. J Neural Transm 111(8):1031-1040.
Abstract: Quantifications of Al, Ca, Cu, Fe, Mg, Mn, Si and Zn were performed in urine, serum, blood and cerebrospinal fluid (CSF) of 26 patients affected by Parkinson's disease (PD) and 13 age-matched controls to ascertain the potential role of biological fluids as markers for this pathology. Analyses were performed by Inductively Coupled Plasma Atomic Emission Spectrometry and Sector Field Inductively Coupled Plasma Mass Spectrometry. The serum oxidant status (SOS) and anti-oxidant capacity (SAC) were also determined. Results showed a decreasing trend for Al in all the fluids of PD patients, with the strongest evidence in serum. Calcium levels in urine, serum and blood of PD patients were significantly higher than in controls. Copper and Mg concentrations were significantly lower in serum of PD patients. Levels of Fe in urine, blood and CSF of patients and controls were dissimilar, with an increase in the first two matrices and a decrease in CSF. No significant difference was found in levels of Mn between patients and controls. Urinary excretion of Si was significantly higher in PD subjects than in controls. No clear difference between Zn levels in the two groups was found for serum, urine or CSF, but an increase in Zn levels in the blood of PD patients was observed. The SOS level in PD was significantly higher while the corresponding SAC was found to be lower in patients than in controls, in line with the hypothesis that oxidative damage is a key factor in the pathogenesis of PD. The results on the whole indicate the involvement of Fe and Zn (increased concentration in blood) as well as of Cu (decreased serum level) in PD. The augmented levels of Ca and Mg in the fluids and of Si in urine of patients may suggest an involuntary intake of these elements during therapy.

Finley JW. 2004. Does environmental exposure to manganese pose a health risk to healthy adults? Nutr Rev 62(4):148-153.
Abstract: Manganese is an essential nutrient that also may be toxic at high concentrations. Subjects chronically exposed to manganese-laden dust in industrial settings develop neuropsychological changes that resemble Parkinson's disease. Manganese has been proposed as an additive to gasoline (as a replacement for the catalytic properties of lead), which has

generated increased research interest in the possible deleterious effects of environmental exposure to manganese. Low-level exposure to manganese has been implicated in neurologic changes, decreased learning ability in school-aged children, and increased propensity for violence in adults. However, a thorough review of the literature shows very weak cause-and-effect relationships that do not justify concern about environmental exposure to manganese for most of the North American population.

Facheris M, Beretta S, Ferrarese C. 2004. Peripheral markers of oxidative stress and excitotoxicity in neurodegenerative disorders: Tools for diagnosis and therapy? Journal of Alzheimers Disease 6(2):177-184.
Abstract: Oxidative stress has been implicated as a common pathogenetic mechanism in neurodegenerative disorders. Central nervous system is particularly exposed to free radical injury, given its high metal content, which can catalyze the formation of oxygen free radicals. and the relatively low content of antioxidant defenses. Indeed, several studies show markers of oxidative damage - lipid peroxidation, protein oxidation, DNA oxidation and glycoxidation markers - in brain areas affected by neurodegenerative disorders. Oxidative stress damage is intimately linked to glutamate neurotoxicity - known as "excitotoxicity". An excessive concentration of extracellular glutamate over-activates ionotropic glutamate receptors, resulting in intracellular calcium overload and a cascade of events leading to neural cell death. In this study we reviewed pathogenetic mechanisms that link oxidative stress and excitotoxicity in three neurodegenerative disorders (Alzheimer's disease, amyotrophic lateral sclerosis and Parkinson's disease) and described peripheral markers of these mechanisms, that may be analyzed in patients as possible diagnostic and therapeutic tools.

Erikson KM, Dobson AW, Dorman DC, Aschner M. 2004. Manganese exposure and induced oxidative stress in the rat brain. Sci Total Environ 334-35(Sp. Iss. Si):409-416.
Abstract: Neurotoxicity linked to excessive brain manganese levels can occur as a result of high level Mn exposures and/or metabolic aberrations (liver disease and decreased biliary excretion). Increased brain manganese levels have been reported to induce oxidative stress, as well as alterations in neurotransmitter metabolism with concurrent neurobehavioral and motor deficits. Two putative mechanisms in which manganese can produce oxidative stress in the brain are: (1) via its oxidation of dopamine, and (2) interference with normal mitochondrial respiration. Measurements of antioxidant species (e.g., glutathione and metallothionein), and the abundance of proteins (enzymes) exquisitely sensitive to oxidation (e.g., glutamine synthetase) have been commonly used as biomarkers of oxidative stress, particularly in rat brain tissue. This paper examines the link between manganese neurotoxicity in the rat brain and common pathways to oxidative stress. (C) 2004 Published by Elsevier B.V.

Dormont D, Ricciardi KG, Tande D, Parain K, Menuel C, Galanaud D, Navarro S, Cornu P, Agid Y, Yelnik J. 2004. Is the subthalamic nucleus hypointense on T2-weighted images? A correlation study using MR imaging and stereotactic atlas data. American Journal of Neuroradiology 25(9):1516-1523.
Abstract: BACKGROUND AND PURPOSE: Although the subthalamic nucleus is the most frequently used target for surgical treatment of Parkinson's disease, the criteria on which it can be identified on T2-weighted images have never been clearly defined. This study was conducted to characterize the precise anatomic distribution of T2-weighted hyposignal in the subthalamic region and to correlate this hyposignal with iron content in the subthalamic nucleus. METHODS: The T2-weighted MR imaging acquisitions of 15 patients with Parkinson's disease were fused with a digitized version of the Schaltenbrand and Wahren anatomic atlas. The MR signal intensity within the anatomic limits of the subthalamic nucleus was evaluated. An anatomic specimen obtained at autopsy was used to evaluate iron content. RESULTS: In all patients, the subthalamic nucleus was hypointense on both sides in the anterior half of the nucleus. At more posterior levels of the

nucleus, hypointensity was less frequently observed (20-80%). Hypointensity was never observed at the most posterior pole. Iron was present in the anteromedial part of the nucleus but absent at the most posterior levels. CONCLUSION. The hypointense signal intensity located lateral to the red nucleus and dorsolateral to the substantia nigra correlates with the presence of iron and corresponds anatomically to the subthalamic nucleus. It can therefore be used as a landmark for electrode implantation in patients with Parkinson's disease. It should, however, be emphasized that although hypointensity was always present in the anterior half of the subthalamic nucleus, the posterior part of the nucleus was not visible in most cases.

Doraiswamy PM, Finefrock AE. 2004. Metals in our minds: therapeutic implications for neurodegenerative disorders. Lancet Neurology 3(7):431-434.
Abstract: Background Abnormal interactions of copper or iron in the brain with metal-binding proteins (such as amyloid-beta peptide [Abeta] or neuromelanin) that lead to oxidative stress have emerged as important potential mechanisms in brain ageing and neurodegenerative disorders. Although a controlled study of desferrioxamine in Alzheimer's disease (AD) had some promising results, concerns about toxicity and brain delivery have limited trials of traditional chelators. The therapeutic significance of metal dysregulation in neurodegenerative disorders has remained difficult to test. Recent developments Clioquinol was identified as a prototype metal-protein-attenuating compound (MPAC). In a blinded and controlled 9 week study of a mouse model of AD, oral clioquinol decreased brain Abeta by 49% without systemic toxicity. The concentrations of copper and zinc in the brain rose by about 15% in mice treated with clioquinol. Two other studies in mice showed that the raising of brain copper concentrations through diet or genetics could lower amyloid load and increase survival. A recent placebo-controlled trial in 36 patients with AD showed that clioquinol (250-750 mg daily) reduced plasma concentrations of Abeta(1-42), raised plasma concentrations of zinc, and-in a subset with moderate dementia-slowed cognitive decline over 24 weeks. Two recent experiments also showed the neuroprotective effects of iron chelation in a mouse model of Parkinson's disease. Where next? The experimental and transgenic-animal studies of metal-protein interactions are convincing but do not provide conclusive answers either about causality or whether this strategy will protect against neurodegeneration in human beings. The finding that clioquinol could modulate plasma concentrations of amyloid and cognition in patients with AD needs to be interpreted cautiously, but is an important first step. Clioquinol was withdrawn because of concerns of its association with subacute myelo-optic neuropathy in Japan; therefore, any additional studies with this drug will likely be small and closely monitored proof-of-concept studies. The development of optimal second-generation MPACs is a desirable goal and may permit greater insights into the significance of metal-protein interactions across several neurodegenerative disorders.

Dopico JG, Diaz JP, Alonso TJ, Hernandez TG, Fuentes RC, Diaz MR. 2004. Extracellular taurine in the substantia nigra: Taurine-glutamate interaction. J Neurosci Res 76(4):528-538.
Abstract: Taurine has been proposed as an inhibitory transmitter in the substantia nigra (SN), but the mechanisms involved in its release and uptake remain practically unexplored. We studied the extracellular pool of taurine in the rat's SN by using microdialysis methods, paying particular attention to the taurine-glutamate (GLU) interaction. Extracellular taurine increased after cell depolarization with high-K+ in a Ca2+-dependent manner, being modified by the local perfusion of GLU, GLU receptor agonists, and zinc. Nigral administration of taurine increased the extracellular concentration of gamma-aminobutyric acid (GABA) and GLU, the transmitters of the two main inputs of the SN. The modification of the glial metabolism with fluocitrate and L-methionine sulfoximine also changed the extracellular concentration of taurine. The complex regulation of the extracellular pool of taurine, its interaction with GABA and GLU, and the involvement of glial cells in its regulation suggest a volume transmission

role for taurine in the SN. (C) 2004 Wiley-Liss, Inc.

Dobson J. 2004. Magnetic Iron Compounds in Neurological DisordersVolume 1012. p 183-192. Redox-Active Metals in Neurological Disorders: Annals of the New York Academy of Sciences.
Abstract: Although iron plays an important role in many aspects of human neurophysiology, it also can be toxic under certain circumstances. Anomalous amounts of iron are known to be associated with most types of neurodegenerative disorders such as Alzheimer's, Parkinson's, and Huntington's diseases. To date, little is known about the specific iron compounds present in this tissue and there is recent evidence to suggest that some forms are magnetic. This raises important questions with regard to the role of magnetic iron compounds in disease initiation and progression and, indeed, the origin of these compounds. This paper reviews recent work on the identification and analysis of magnetic iron compounds associated with neurological disorders.

Dobson AW, Erikson KM, Aschner M. 2004. Manganese NeurotoxicityVolume 1012. p 115-128. Redox-Active Metals in Neurological Disorders: Annals of the New York Academy of Sciences.
Abstract: Manganese is an essential trace element and it is required for many ubiquitous enzymatic reactions. While manganese deficiency rarely occurs in humans, manganese toxicity is known to occur in certain occupational settings through inhalation of manganese-containing dust. The brain is particularly susceptible to this excess manganese, and accumulation there can cause a neurodegenerative disorder known as manganism. Characteristics of this disease are described as Parkinson-like symptoms. The similarities between the two disorders can be partially explained by the fact that the basal ganglia accumulate most of the excess manganese compared with other brain regions in manganism, and dysfunction in the basal ganglia is also the etiology of Parkinson's disease. It has been proposed that populations already at heightened risk for neurodegeneration may also be more susceptible to manganese neurotoxicity, which highlights the importance of investigating the human health effects of using the controversial compound, methylcyclopentadienyl manganese tricarbonyl (MMT), in gasoline to increase octane. The mechanisms by which increased manganese levels can cause neuronal dysfunction and death are yet to be elucidated. However, oxidative stress generated through mitochondrial perturbation may be a key event in the demise of the affected central nervous system cells. Our studies with primary astrocyte cultures have revealed that they are a critical component in the battery of defenses against manganese-induced neurotoxicity. Additionally, evidence for the role of oxidative stress in the progression of manganism is reviewed here.

Diaz-Veliz G, Mora S, Gomez P, Dossi MT, Montiel J, Arriagada C, Aboitiz F, Segura-Aguilar J. 2004. Behavioral effects of manganese injected in the rat substantia nigra are potentiated by dicumarol, a DT-diaphorase inhibitor. Pharmacol Biochem Behav 77(2):245-251.
Abstract: The purpose of this study was to evaluate the contribution of DT-diaphorase inhibition to in vivo neurodegenerative effects of dopamine (DA) oxidation to the corresponding o-quinones. The neurotoxicity to nigrostriatal DA neurons was induced by injection of manganese pyrophosphate (Mn3+) complex as a prooxidizing agent alone or together with the DT-diaphorase inhibitor dicumarol into the right rat substantia nigra. The behavioral effects were compared with those induced after selective lesions of dopaminergic neurons with 6-hydroxydopamine (6-OHDA). Intranigral injection of Mn3+ and Mn3+ plus dicumarol produced significant impairment in motor behavior compared with control animals. However, the effect seen in the Mn3+ plus dicumarol injected group was significantly more severe than that observed in the Mn3+ alone injected group. In motor activity and rearing behavior, the simultaneous injection of Mn3+ plus dicumarol produced a 6-OHDA-like impairment. Similar effects were observed in the acquisition of a conditioned avoidance response (CAR). Dicumarol significantly impaired avoidance conditioning although

without affecting the motor behavior. The behavioral effects were correlated to the extent of striatal tyrosine hydroxylase (TH)-positive fiber loss. Rats receiving unilateral intranigral Mn3+ and Mn3+ plus dicumarol injections exhibited a significant reduction in nigrostriatal TH-positive fiber density in medial forebrain bundle compared with the contralateral noninjected side. In conclusion, this study provides evidence that the neurotoxicity of Mn3+ in vivo is potentiated by DT-diaphorase inhibition, suggesting that this enzyme could play a neuroprotective role in the nigrostriatal DA systems. (C) 2003 Elsevier Inc. All rights reserved.

Di JW, Bi SP. 2004. Aluminum facilitation of the iron-mediated oxidation of DOPA to melanin. Analytical Sciences 20(4):629-634.
Abstract: Aluminum, a trivalent cation unable to undergo redox reactions, is shown to faciliate iron-initiated DOPA oxidation in the melanin pathway under acidic condition of pH 5.5, which is a favored medium for aluminum facilitation of iron-induced lipid peroxidation. In the process of oxidation of DOPA to melanin in the presence of the metal ions, Fe3+ and H2O2 oxidize DOPA to dopachrome (DC), then Al3+ catalyzes the conversion of DC to 5,6-dihydroxyindole (DHI) and finally Fe3+ oxidizes DHI to indole-5,6-quinone (IQ), which polymerizes immediately to melanochrome and melanin. The reactions involve the intermediate complexes of metal ions and DOPA or its derivative. The present results indicate that aluminum can enhance the oxidative stress on iron-mediated DOPA oxidation in melanin pathway under acidic condition through the cooperation of iron and aluminum ions.

Dhanasekaran A, Kotamraju S, Kalivendi SV, Matsunaga T, Shang T, Keszler A, Joseph J, Kalyanaraman B. 2004. Supplementation of endothelial cells with mitochondria-targeted antioxidants inhibit peroxide-induced mitochondrial iron uptake, oxidative damage, and apoptosis. J Biol Chem 279(36): 37575-37587.
Abstract: The mitochondria-targeted drugs mitoquinone (Mito-Q) and mitovitamin E (MitoVit-E) are a new class of antioxidants containing the triphenylphosphonium cation moiety that facilitates drug accumulation in mitochondria. In this study, Mito-Q (ubiquinone attached to a triphenylphosphonium cation) and MitoVit-E (vitamin E attached to a triphenylphosphonium cation) were used. The aim of this study was to test the hypothesis that mitochondria-targeted antioxidants inhibit peroxide-induced oxidative stress and apoptosis in bovine aortic endothelial cells (BAEC) through enhanced scavenging of mitochondrial reactive oxygen species, thereby blocking reactive oxygen species-induced transferrin receptor (TfR)-mediated iron uptake into mitochondria. Glucose/glucose oxidase-induced oxidative stress in BAECs was monitored by oxidation of dichlorodihydrofluorescein that was catalyzed by both intracellular H2O2 and transferrin iron transported into cells. Pretreatment of BAECs with Mito-Q (1 muM) and MitoVit-E (1 muM) but not untargeted antioxidants (e. g. vitamin E) significantly abrogated H2O2- and lipid peroxide-induced 2', 7'-dichlorofluorescein fluorescence and protein oxidation. Mitochondria-targeted antioxidants inhibit cytochrome c release, caspase-3 activation, and DNA fragmentation. Mito-Q and MitoVit-E inhibited H2O2- and lipid peroxide-induced inactivation of complex I and aconitase, TfR overexpression, and mitochondrial uptake of Fe-55, while restoring the mitochondrial membrane potential and proteasomal activity. We conclude that Mito-Q or MitoVit-E supplementation of endothelial cells mitigates peroxide-mediated oxidant stress and maintains proteasomal function, resulting in the overall inhibition of TfR-dependent iron uptake and apoptosis.

Deplazes J, Schobel K, Hochstrasser H, Bauer P, Walter U, Behnke S, Spiegel J, Becker G, Riess O, Berg D. 2004. Screening for mutations of the IRP2 gene in Parkinson's disease patients with hyperechogenicity of the substantia nigra. J Neural Transm 111(4):515-521.
Abstract: IRP2 plays an important role in brain iron metabolism. We recently identified an increased amount of iron in patients with Parkinson's disease (PD) and hyperchogenicity of the substantia nigra (SN). Therefore,

the IRP2 gene was screened for mutations in 176 PD patients with increased echogenicity of the SN. We identified one non-synonymous polymorphism (I888V) in exon 21 and a -88C > T polymorphism in the promoter region of IRP2 at similar frequencies in patients and controls without increased SN iron levels. In one patient a -74C > T variation was found which was not present in the control group. Our data indicate that mutations in the IRP2 gene are not a common cause of PD associated with SN iron accumulation.

Del Rio MJ, Velez-Pardo C. 2004. Transition metal-induced apoptosis in generation, mitochondria dysfunction lymphocytes via hydroxyl radical, and caspase-3 activation: An in vitro model for neurodegeneration. Arch Med Res 35(3):185-193.
Abstract: Background. Redox transition metals have been implicated as crucial players in pathogenesis of neurodegenerative diseases. Intracellular signaling mechanism(s) responsible C for oxidative stress and death in single-cell model exposed to metals has not yet been fully elucidated. The objective of the study was to determine the mechanism by which metals induced apoptosis in human peripheral blood lymphocytes (PBL). Methods. PBL were exposed to 50, 100, 250, 500, and 1,000 muM (Fe2+), (Mn2+), (Cu2+), and (Zn2+)-(SO4). Apoptotic/necrotic morphology was assessed with acridine orange/ethidium bromide staining. Further evaluations comprised production of H2O2, generation of hydroxyl radical (.OH), disruption of mitochondrial transmembrane potential (DeltaPsi(m)), caspase-3 activation, and activation of NF-kappaB and p53 transcriptional factors. Results. Morphologic analysis showed that 500 muM provoked maximal percentage of apoptosis (22-30% AO/EB) and minimal necrosis (3-7%), whereas low concentrations were innocuous but 1,000 muM induced mainly necrosis (>40% AO/EB). Metals generated both H2O2, and (.OH) by Fenton reaction. Hydroxyl scavengers protected PBL from metal-induced apoptosis. All metals induced mitochondrial depolarization (17-62% nonfluorescent cells) and activated caspase-3 concomitantly with apoptotic morphology (25-32% AO/EB) at 24 h, and neither NF-kappaB nor p53 transcription factor showed activation. Conclusions. This study provides evidence that redox-active (Fe2+), (Mn2+), (Cu2+), and (Zn2+) ion-induced apoptosis in PBL by (H2O2)/(.OH) generation, resulting in mitochondria depolarization, caspase-3 activation, and nuclear fragmentation independent of NF-kappaB and p53 transcription factors activation. Our data highlight the potential use of lymphocytes as a model to screen antioxidant strategies designed to remove H2O2/.OH associated with metal-catalyzed reactions in neurodegenerative disorders. (C) 2004 IMSS. Published by Elsevier Inc.

Del Rio MJ, Moreno S, Garcia-Ospina G, Buritica O, Uribe CS, Lopera F, Velez-Pardo C. 2004. Autosomal recessive juvenile parkinsonism Cys212Tyr mutation in parkin renders lymphocytes susceptible to dopamine- and iron-mediated apoptosis. Mov Disord 19(3):324-330.
Abstract: Mutations in parkin are implicated in the pathogenesis of autosomal recessive juvenile parkinsonism (AR-JP) disease. We show that homozygote Cys212Tyr parkin mutation in AR-JP patients renders lymphocytes sensitive to dopamine, iron and hydrogen peroxide stimuli. Indeed, dopamine-induced apoptosis by four alternative mechanisms converging on caspase-3 activation and apoptotic morphology: (1) NF-kappaB-dependent pathway; mitochondrial dysfunction either by (2) H2O2 or (3) hydroxyl exposure and (4) increase of unfolded-protein stress. We also demonstrate that 17beta-estradiol and testosterone prevent homozygote lymphocytes from oxidative stressors-evoked apoptosis. These results may contribute to understanding the relationship between genetic and environmental factors and iron in AR-JP. (C) 2004 Movement Disorder Society.

Deane R, Zheng W, Zlokovic BV. 2004. Brain capillary endothelium and choroid plexus epithelium regulate transport of transferrin-bound and free iron into the rat brain. J Neurochem 88(4):813-820.
Abstract: Iron transport into the CNS is still not. completely understood.

Using a brain perfusion technique in rats, we have shown a significant brain capillary uptake of circulating transferrin (Tf)-bound and free Fe-59 (1 nM) at rates of 136 +/- 26 and 182 +/- 23 muL/g/min, respectively, while their respective transport rates into brain parenchyma were 1.68 +/- 0.56 and 1.52 +/- 0.48 muL/g/min. Regional Tf receptor density (B-max) in brain endothelium determined with I-125-holo-Tf correlated well with Fe-59-Tf regional brain uptake rates reflecting significant vascular association of iron. Tf-bound and free circulating Fe-59 were sequestered by the choroid plexus and transported into the CSF at low rates of 0.17 +/- 0.01 and 0.09 +/- 0.02 muL/min/g, respectively, consistent with a 10-fold brain-CSF concentration gradient for Fe-59, Tf-bound or free. We conclude that transport of circulating Tf-bound and free iron could be equally important for its delivery to the CNS. Moreover, data suggest that entry of Tf-bound and free iron into the CNS is determined by (i) its initial sequestration by brain capillaries and choroid plexus, and (ii) subsequent controlled and slow release from vascular structures into brain interstitial fluid and CSF.

De La Fuente-Fernandez R. 2004. Portal-systemic shunts, manganese, and parkinsonism. J Neurol Neurosurg Psychiatry 75(7):1081.

Crossgrove J, Zheng W. 2004. Manganese toxicity upon overexposure. NMR Biomed 17(8):544-553.
Abstract: Manganese (Mn) is a required element and a metabolic byproduct of the contrast agent mangafodipir trisodium (MnDPDP). The Mn released from MnDPDP is initially sequestered by the liver for first-pass elimination. which allows an enhanced contrast for diagnostic imaging. The administration of intravenous Mn impacts its homeostatic balance in the human body and can lead to toxicity. Human Mn deficiency has been reported in patients oil parenteral nutrition and in micronutrient studies. Mn toxicity has been reported through occupational (e.g. welder) and dietary overexposure and is evidenced primarily in the central nervous system, although lung. cardiac, liver. reproductive and fetal toxicity have been noted. Mn neurotoxicity results from all accumulation of the metal in brain tissue and results in a progressive disorder of the extrapyramidal system which is similar to Parkinson's disease. In order for Mn to distribute from blood into brain tissue, it must cross either the blood-brain barrier (BBB) or the blood-cerebrospinal fluid barrier (BCB). Brain import, with no evidence of export, would lead to brain Mn accumulation and neurotoxicity. The mechanism for the neuro-degenerative damage specific to select brain regions is not clearly understood. Disturbances in iron homeostasis and the valence state of Mn have been implicated as key factors in contributing to Mn toxicity. Chelation therapy with EDTA and supplementation with levodopa are the current treatment options, which are mildly and transiently efficacious. In conclusion. repeated administration of Mn Or compounds that readily release Mn. may increase the risk of Mn-induced toxicity. Copyright (C) 2004 John Wiley Soils. Ltd.

Crichton RR, Ward RJ. 2004. Iron Chelators and Their Therapeutic PotentialVolume 41. p 185-219. Metal Ions in Biological Systems, Vol 41: Metal Ions and Their Complexes in Medication: Metal Ions in Biological Systems.

Costello DJ, Walsh SL, Harrington HJ, Walsh CH. 2004. Concurrent hereditary haemochromatosis and idiopathic Parkinson's disease: a case report series. J Neurol Neurosurg Psychiatry 75(4):631-633.
Abstract: Hereditary haemochromatosis (HH) is a genetic disorder in which abnormal iron handling leads to excessive iron accumulation in systemic tissues. Magnetic resonance imaging studies suggest excess iron deposition in the basal ganglia of patients with HH. The symptoms of neurological complications of HH include cognitive decline, gait difficulties, cerebellar ataxia, and extrapyramidal dysfunction, but idiopathic Parkinson's disease, in which brain iron deposition is normal, has not been reported. We describe four patients with concurrent HH and IPD. Although three of the cases had risk factors for cerebrovascular and cardiovascular disease, computed tomography did not show ischaemic changes in the basal

ganglia. We speculate that in these cases, abnormal deposition of iron in the basal ganglia induced the symptoms of IPD.

Connor JR, Wang XS, Patton SM, Menzies SL, Troncoso JC, Earley CJ, Allen RP. 2004. Decreased transferrin receptor expression by neuromelanin cells in restless legs syndrome. Neurology 62(9):1563-1567.
Abstract: Background: Restless legs syndrome (RLS) is a sensory-movement disorder affecting 5 to 10% of the population. Its etiology is unknown, but MRI analyses and immunohistochemical studies on autopsy tissue suggest the substantia nigra (SN) of patients with RLS has subnormal amounts of iron. Methods: Neuromelanin cells from the SN of four RLS and four control brains were isolated by laser capture microdissection, and a profile of iron-management protein expression was obtained by immunoblot analysis. Binding assays for iron regulatory protein activity were performed on cell homogenates. Results: Ferritin, divalent metal transporter 1, ferroportin, and transferrin receptor (TfR) were decreased in RLS neuromelanin cells compared with control. Transferrin was increased in RLS neuromelanin cells. This protein profile in RLS neuromelanin cells is consistent with iron deficiency with the exception that TfR expression was decreased rather than increased. The concentration and activity of the iron regulatory proteins (IRP1 and IRP2) were analyzed to determine whether there was a functional deficit in the post-transcriptional regulatory mechanism for TfR expression. Total IRP activity, IRP1 activity, and IRP1 protein levels were decreased in RLS, but total IRP2 protein levels were not decreased in RLS. Conclusion: Restless legs syndrome may result from a defect in iron regulatory protein 1 in neuromelanin cells that promotes destabilization of the transferrin receptor mRNA, leading to cellular iron deficiency.

Cheng SY, Trombetta LD. 2004. The induction of amyloid precursor protein and alpha-synuclein in rat hippocarnpal astrocytes by diethyldithiocarbamate and copper with or without glutathione. Toxicol Lett 146(2):139-149.
Abstract: alpha-Synuclein is the major component of Lewy bodies. Its aggregation can be accelerated by copper, iron, or beta-amyloid (Abeta) and has been thought to provide a nucleation center during the formation of amyloid plaques. The main structural component of amyloid plaque is Abeta, which is derived from a larger protein, amyloid precursor protein (APP). Xenobiotics have been implicated in the etiology of the neurodegenerative disease. Mechanisms of diethyldithiocarbamate (DDC) neurotoxicity involve copper chelation and interactions with SH groups resulting in oxidative stress. In this study, rat hippocampal astrocytes were treated with DDC (75 muM), CuCl2 (0.2 muM), or DDC (75 muM) Plus CuCl2 (0.2 muM) for 1 h. Cells were allowed to recover with or without 10 mM GSH. Results showed an increase of APP and alpha-synuclein production occurring in a time-dependent manner. At 4 h post-treatment, cells contained small positively stained material deposited throughout the cytosol for APP and by 8 h post-treatment increases were seen in both APP and alpha-synuclein. Immunoblots supported immunocytochemical results. Glutathione (GSH) decreased the accumulation of these proteins at 8 h post-treatment. (C) 2003 Elsevier Ireland Ltd. All rights reserved.

Chauhan A, Chauhan V, Brown WT, Cohen I. 2004. Oxidative stress in autism: Increased lipid peroxidation and reduced serum levels of ceruloplasmin and transferrin - the antioxidant proteins. Life Sci 75(21):2539-2549.
Abstract: Autism is a neurological disorder of childhood with poorly understood etiology and pathology. We compared lipid peroxidation status in the plasma of children with autism, and their developmentally normal non-autistic siblings by quantifying the levels of malonyldialdehyde, an end product of fatty acid oxidation. Lipid peroxidation was found to be elevated in autism indicating that oxidative stress is increased in this disease. Levels of major antioxidant proteins namely, transferrin (iron-binding protein) and ceruloplasmin (copper-binding protein) in the serum, were significantly reduced in autistic children as compared to their developmentally normal non-autistic siblings. A striking correlation was observed between reduced levels of these proteins and loss of previously acquired language skills in

children with autism. These results indicate altered regulation of transferrin and ceruloplasmin in autistic children who lose acquired language skills. It is suggested that such changes may lead to abnormal iron and copper metabolism in autism, and that increased oxidative stress may have pathological role in autism. (C) 2004 Elsevier Inc. All rights reserved.

Carod-Artal FJ, Vargas AP, Marinho PB, Fernandes-Silva TV, Portugal D. 2004. Tourettism, hemiballism and juvenile Parkinsonism: Expanding the clinical spectrum of the neurodegeneration associated to pantothenate kinase deficiency (Hallervorden-Spatz syndrome). Rev Neurol 38(4):327-331.
Abstract: Introduction. Pantothenate kinase deficiency (Hallervorden-Spatz syndrome, HSS) triggers cerebral neurodegeneration with iron deposition in the basal ganglia. The classical form has an early onset in infancy, a progressive course, the presence of extrapyramidal symptoms (dystonia, chorea, rigidity) and pigmentary retinitis. There are atypical late onset forms with predominance of symptoms of Parkinsonism and dementia, which progress slowly and course somewhat less progressively. Case report. We describe three patients with HSS and an atypical presentation, with onset during the second decade of life. In all cases magnetic resonance imaging showed areas of hyposignal in T-2 sequences in medial globus pallidus, with central hypersignal, which gave rise to a tiger's eye image. Other aetiologies, such as Wilson's disease, gangliosidosis GM1, hypoprebetalipoproteinemia, hexosaminidase A deficiency, aminoacidurias and infantile Huntingdon's chorea, were precluded. In the 20-year-old male the initial manifestations at the age of 17 were superposed over Gilles de la Tourette syndrome, with complex motor and vocal tics, palilalia, behavioural disorders and postural instability. The 13-year-old patient resented symptoms of chorea, hemiballic movements and dystonia in the lower limbs, which limited walking at the age of 12. The 28-year-old female patient presented a progressive rigid-akinetic syndrome, with dementia and partial response to levodopa. Conclusions. The clinical spectrum of HSS is broad and its differential diagnosis must include hemiballism, Tourette syndrome and juvenile Parkinsonism.

Cardoso SM, Santana I, Swerdlow RH, Oliveira CR. 2004. Mitochondria dysfunction of Alzheimer's disease cybrids enhances A beta toxicity. J Neurochem 89(6):1417-1426.
Abstract: Alzheimer's disease (AD) brain reveals high rates of oxygen consumption and oxidative stress, altered antioxidant defences, increased oxidized polyunsaturated fatty acids, and elevated transition metal ions. Mitochondrial dysfunction in AD is perhaps relevant to these observations, as such may contribute to neurodegenerative cell death through the formation of reactive oxygen species (ROS) and the release of molecules that initiate programmed cell death pathways. In this study, we analyzed the effects of beta-amyloid peptide (Abeta) on human teratocarcinoma (NT2) cells expressing endogenous mitochondrial DNA (mtDNA), mtDNA from AD subjects (AD cybrids), and mtDNA from age-matched control subjects (control cybrids). In addition to finding reduced cytochrome oxidase activity, elevated ROS, and reduced ATP levels in the AD cybrids, when these cell lines were exposed to Abeta 1-40 we observed excessive mitochondrial membrane potential depolarization, increased cytoplasmic cytochrome c, and elevated caspase-3 activity. When exposed to Abeta, events associated with programmed cell death are activated in AD NT2 cybrids to a greater extent than they are in control cybrids or the native NT2 cell line, suggesting a role for mtDNA-derived mitochondrial dysfunction in AD degeneration.

Capili AD, Edghill EL, Wu K, Borden KLB. 2004. Structure of the C-terminal RING finger from a RING-IBR-RING/TRIAD motif reveals a novel zinc-binding domain distinct from a RING. J Mol Biol 340(5):1117-1129.
Abstract: The really interesting new gene (RING) family of proteins contains over 400 members with diverse physiological functions. A subset of these domains is found in the context of the RING-IBR-RING/TRIAD motifs which function as E3 ubiquitin ligases. Our sequence analysis of the C-terminal RING (RING2) from this motif show that several metal ligating

and hydrophobic residues critical for the formation of a classical RING 2 cross-brace structure are not present. Thus, we determined the structure of the RING2 from the RING-IBR-RING motif of HHARI and showed.. that RING2 has a completely distinct topology from classical RINGs. Notably, RING2 binds only one zinc atom per monomer rather than two and uses a different hydrophobic network to that of classical RINGs. Additionally, this RING2 topology is novel, bearing slight resemblance to zinc-ribbon motifs around the zinc site and is different from the topologies of the zinc binding sites found in RING and PHDs. We demonstrate that RING2 acts as an E3 ligase in vitro and using mutational analysis deduce the structural features required for this activity. Further, mutations in the RING-IBR-RING of Parkin cause a rare form of Parkinsonism and these studies provide an explanation for those mutations that occur in its RING2. From a comparison of the RING2 structure with those reported for RINGs, we infer sequence determinants that allow discrimination between RING2 and RING domains at the sequence analysis level. (C) 2004 Elsevier Ltd. All rights reserved.

Capanni C, Taddei N, Gabrielli S, Messori L, Orioli P, Chiti F, Stefani M, Ramponi G. 2004. Investigation of the effects of copper ions on protein aggregation using a model system. Cell Mol Life Sci 61(7-8):982-991.
Abstract: Protein aggregation is a notable feature of various human disorders, including Parkinson's disease, Alzheimer's disease and many others systemic amyloidoses. An increasing number of observations in vitro suggest that transition metals are able to accelerate the aggregation process of several proteins found in pathological deposits, e.g. alpha-synuclein, amyloid beta (Abeta) peptide, beta(2)-microglobulin and fragments of the prion protein. Here we report the effects of metal ions on the aggregation rate of human muscle acylphosphatase, a suitable model system for aggregation studies in vitro. Among the different species tested, Cu2+ produced the most remarkable acceleration of aggregation, the rate of the process being 2.5-fold higher in the presence of 0.1 mM metal concentration. Data reported in the literature suggest the possible role played by histidine residues or negatively charged clusters present in the amino acid sequence in Cu2+-mediated aggregation of pathological proteins. Acylphosphatase does not contain histidine residues and is a basic protein. A number of histidine-containing mutational variants of acylphosphatase were produced to evaluate the importance of histidine in the aggregation process. The Cu2+-induced acceleration of aggregation was not significantly altered in the protein variants. The different aggregation rates shown by each variant were entirely explained by the changes of hydrophobicity or propensity to form a beta structure introduced by the point mutation. The effect of Cu2+ on acylphosphatase aggregation cannot therefore be attributed to the specific factors usually invoked in the aggregation of pathological proteins. The effect, rather, seems to be a general related to the chemistry of the polypeptide backbone and could represent an additional deleterious factor resulting from the alteration of the homeostasis of metal ions in cells.

Cairns NJ, Lee VMY, Trojanowski JQ. 2004. The cytoskeleton in neurodegenerative diseases. J Pathol 204(4):438-449.
Abstract: Abundant abnormal aggregates of cytoskeletal proteins are neuropathological signatures of many neurodegenerative diseases that are broadly classified by filamentous aggregates of neuronal intermediate filament (IF) proteins, or by inclusions containing the microtubule-associated protein (MAP) tau. The discovery of mutations in neuronal IF and tau genes firmly establishes the importance of neuronal IF proteins and tau in the pathogenesis of neurodegenerative diseases. Multiple IF gene mutations are pathogenic for Charcot-Marie-Tooth (CMT) disease and amyotrophic lateral sclerosis (ALS) - in addition to those in the copper/zinc superoxide dismutase-1 (SOD1) gene. Tau gene mutations are pathogenic for frontotemporal dementia with parkinsonism linked to chromosome 17 (FTDP-17), and tau polymorphisms are genetic risk factors for sporadic progressive supranuclear palsy (PSP) and corticobasal degeneration (CBD). Thus, IF and tau abnormalities are linked directly to the aetiology and pathogenesis of neurodegenerative diseases. In vitro and transgenic

animal models are being used to demonstrate that different mutations impair protein function, promote tau fibrilization, or perturb tau gene splicing, leading to aberrant and distinct tau aggregates. For recognition of these disorders at neuropathological examination, immunohistochemistry is needed, and this may be combined with biochemistry and molecular genetics to properly determine the nosology of a particular case. As reviewed here, the identification of molecular genetic defects and biochemical alterations in cytoskeletal proteins of human neurodegenerative diseases has facilitated experimental studies and will promote the development of assays of molecules which inhibit abnormal neuronal IF and tau protein inclusions. Copyright (C) 2004 Pathological Society of Great Britain and Ireland. Published by John Wiley Sons, Ltd.

Brown GC, Borutaite V. 2004. Inhibition of mitochondrial respiratory complex I by nitric oxide, peroxynitrite and S-nitrosothiols. Biochimica Et Biophysica Acta-Bioenergetics 1658(1-2. Sp. Iss. Si):44-49.
Abstract: NO or its derivatives (reactive nitrogen species, RNS) inhibit mitochondrial complex I by several different mechanisms that are not well characterised. There is an inactivation by NO, peroxynitrite and S-nitrosothiols that is reversible by light or reduced thiols, and therefore may be due to S-nitrosation or Fe-nitrosylation of the complex. There is also an irreversible inhibition by peroxynitrite, other oxidants and high levels of NO, which may be due to tyrosine nitration, oxidation of residues or damage of iron sulfur centres. Inactivation of complex I by NO or RNS is seen in cells or tissues expressing iNOS, and may be relevant to inflammatory pathologies, such as septic shock and Parkinson's disease. (C) 2004 Elsevier B.V. All rights reserved.

Bridge MH, Williams E, Lyons MEG, Tipton KF, Linert W. 2004. Electrochemical investigation into the redox activity of Fe(II)/Fe(III) in the presence of nicotine and possible relations to neurodegenerative diseases. Biochimica Et Biophysica Acta-Molecular Basis of Disease 1690(1):77-84.
Abstract: The biological relevance of Fe(II)/Fe(III) is becoming evermore apparent, especially in relation to its potential role in the progression of neurodegenerative diseases such as Parkinson's and Alzheimer's disease. The reported relationship between smoking and a reduced incidence of neurodegenerative disorders prompted this work. In order to investigate whether nicotine can interact with iron, we have studied the electrochemical behaviour of a Fe(II)/Fe(III) redox couple in the presence of nicotine. Solubility issues and lack of available nonreacting salts of nicotine necessitated studies being conducted at low pH values. Cyclic voltammetry experiments revealed a definite alteration in the electrochemical behaviour of the Fe(II)/Fe(III) redox couple suggesting the capability of nicotine to complex with free iron and, hence, reduce its reactivity. This is evident from a slower rate of heterogeneous electron transfer, k(s), and a shift from reversible to quasi-reversible behaviour, as characterised from the diffusion coefficient (D), the full width half maximum (FWHM), DeltaE(p) and E-f. Additional complexation titrations, pH ranging from 1 to 7, confirm a weak complexation reaction occurring between Fe(III) and nicotine. (C) 2004 Elsevier B.V. All rights reserved.

Bossy-Wetzel E, Schwarzenbacher R, Lipton SA . 2004. Molecular pathways to neurodegeneration. Nat Med :S2-S9.
Abstract: The molecular bases underlying the pathogenesis of neurodegenerative diseases are gradually being disclosed. One problem that investigators face is distinguishing primary from secondary events. Rare, inherited mutations causing familial forms of these disorders have provided important insights into the molecular networks implicated in disease pathogenesis. Increasing evidence indicates that accumulation of aberrant or misfolded proteins, protofibril formation, ubiquitin-proteasome system dysfunction, excitotoxic insult, oxidative and nitrosative stress, mitochondrial injury, synaptic failure, altered metal homeostasis and failure of axonal and dendritic transport represent unifying events in many slowly progressive neurodegenerative disorders.

Bossy-Wetzel E, Schwarzenbacher R, Lipton SA . 2004. Molecular pathways to neurodegeneration. Nature Reviews Neuroscience :S2-S9.
Abstract: The molecular bases underlying the pathogenesis of neurodegenerative diseases are gradually being disclosed. One problem that investigators face is distinguishing primary from secondary events. Rare, inherited mutations causing familial forms of these disorders have provided important insights into the molecular networks implicated in disease pathogenesis. Increasing evidence indicates that accumulation of aberrant or misfolded proteins, protofibril formation, ubiquitin-proteasome system dysfunction, excitotoxic insult, oxidative and nitrosative stress, mitochondrial injury, synaptic failure, altered metal homeostasis and failure of axonal and dendritic transport represent unifying events in many slowly progressive neurodegenerative disorders.

Borlongan CV, Wang Y, Su TP. 2004. Delta opioid peptide (D-Ala 2, D-Leu 5) enkephalin: Linking hibernation and neuroprotection . Front Biosci 9:3392-3398.
Abstract: Hibernation is a potential protective strategy for the peripheral, as well as for the central nervous system. A protein factor termed hibernation induction trigger (HIT) was found to induce hibernation in summer-active ground squirrels. Purification of HIT yielded an 88-kD peptide that is enriched in winter hibernators. Partial sequence of the 88-kD protein indicates that it may be related to the inhibitor of metalloproteinase. Using opioid receptor antagonists to elucidate the mechanisms of HIT, it was found that HIT targeted the delta opioid receptors. Indeed, delta opioid (D-Ala 2, D-Leu 5) enkephalin (DADLE) was shown to induce hibernation. Specifically, HIT and DADLE were found to prolong survival of peripheral organs, such as the lung, the heart, liver, and kidney preserved en bloc or as a single preparation. In addition, DADLE has been recently demonstrated to promote survival of neurons in the central nervous system. Exposure to DADLE dose-dependently enhanced cell viability of cultured primary rat fetal dopaminergic cells. Subsequent transplantation of these DADLE-treated dopaminergic cells into the Parkinsonian rat brain resulted in a two-fold increase in surviving grafted cells. Interestingly, delivery of DADLE alone protected against dopaminergic depletion in a rodent model of Parkinson's disease. Similarly, DADLE blocked and reversed the dopaminergic terminal damage induced by methamphetamine (METH). Such neuroprotective effects of DADLE against METH neurotoxicity was accompanied by attenuation of mRNA expressions of a tumor necrosis factor p53 and an immediate early gene c-fos. In parallel to these beneficial effects of DADLE on the dopaminergic system, DADLE also ameliorated the neuronal damage induced by ischemia-reperfusion following a transient middle cerebral artery occlusion. In vitro replication of this ischemia cell death by serum-deprivation of PC12 cells revealed that DADLE exerted neuroprotection in a naltrexone-sensitive manner. These results taken together suggest that DADLE stands as a novel therapeutic agent. In this review paper, we present laboratory evidence supporting the use of DADLE for protection of peripheral and central nervous system.

Bocca B, Alimonti A, Petrucci F, Violante N, Sancesario G, Forte G, Senofonte O. 2004. Quantification of trace elements by sector field inductively coupled plasma mass spectrometry in urine, serum, blood and cerebrospinal fluid of patients with Parkinson's disease. Spectrochimica Acta Part B-Atomic Spectroscopy 59(4):559-566.
Abstract: To assess whether levels of trace metals and oxidative species are involved in Parkinson's disease (PD), At, Be, Cd, Co, Cr, Ha, Mn, Ni, Pb and V were measured in urine, serum, blood and cerebrospinal fluid (CSF) and serum peroxides and antioxidant capacity were determined in 26 patients with PD and 13 control subjects. The quantification of metals was based on the 1 + 4 water dilution of CSF, serum and urine, the acid-assisted microwave digestion under atmospheric pressure of blood and final determination by sector field inductively coupled plasma mass spectrometry (SF-ICP-MS). Results indicated a significant increase of Pb and V concentrations in blood and urine (P less than or equal to 0.03, in

both cases) related to the disease. Parkinson disease also seemed to be closely associated (P less than or equal to 0.003) with a reduction in levels of Al, Cd, Hg and Pb in serum and of Cd, Co, Cr, Hg, Pb in CSF As regards Mn, a lower mean concentration was found in the CSF and whole blood of PD patients than in control group, although this trend was not statistically significant. Levels of peroxides were also increased (P less than or equal to 0.001), while antioxidant capacity was lower (P less than or equal to 0.002) in PD patients than in controls. (C) 2004 Published by Elsevier B.V.

Blandini F, Cosentino M, Mangiagalli A, Marino F, Samuele A, Rasini E, Fancellu R, Tassorelli C, Pacchetti C, Martignoni E, Riboldazzi G, Calandrella D, Lecchini S, Frigo G, Nappi G. 2004. Modifications of apoptosis-related protein levels in lymphocytes of patients with Parkinson's disease. The effect of dopaminergic treatment. J Neural Transm 111(8):1017-1030.
Abstract: In this study, we investigated whether changes in the regulatory mechanisms of apoptosis and oxidative stress may be detected, peripherally, in patients with Parkinson's disease (PD). For this purpose, we measured caspase-3 activity, Bcl-2 concentrations, peripheral benzodiazepine receptor (PBR) expression and Cu/Zn superoxide dismutase (SOD) concentrations in lymphocytes of untreated PD patients, patients treated only with L-Dopa or with L-Dopa and dopamine agonists and healthy volunteers. Caspase-3 activity was significantly increased in all PD patient groups. Patients treated with L-Dopa and dopamine agonists showed the lowest values of Bcl-2, coupled with the highest density of PBRs, while increased levels of Cu/Zn SOD were found in the group under monotherapy with L-Dopa. We also found, in PD patients, clear, negative correlations between Bcl-2 levels and both duration and severity of the disease. Our findings point to the existence of changes in the regulatory mechanisms of apoptosis in PD patients - observable outside the central nervous system - which seem to be modulated by the pharmacological treatment with dopaminergic agents.

Beuter A, Lambert G, Macgibbon B. 2004. Quantifying postural tremor in workers exposed to low levels of manganese. J Neurosci Methods 139(2):247-255.
Abstract: The aim of this study was: (1) To determine the minimum number of characteristics necessary to discriminate between postural tremor recorded in control subjects (CO), in subjects exposed to manganese (MN), and in patients with Parkinson's disease (PD), and (2) to examine the continuum of changes between the three groups examined. Workers previously exposed to Mn (n = 10), patients with PD (n = 10), and control subjects (CO) (n = 11) underwent a clinical examination. Blood Mn was measured at the end of exposure time for the MN group and 12 months later at the beginning of the experiment for all groups. Postural tremor with visual feedback was recorded in the index finger with a laser system. Statistical criteria were used to reduce computed tremor characteristics to a minimal set of reliable discriminating variables. Two variables were retained namely corrected wobble (CW), describing the morphology of the tremor oscillations, and variability ratio (VR), describing proportional power of tremor. Both variables had an overall correct classification rate of 77.4%. Blood Mn levels at the time of the experiment were similar for all groups and had insignificant correlation with tremor variables. However, blood Mn levels in workers which were also measured at the end of exposure time (i.e., 12 months before) showed significant correlation (Spearman's rank coefficient) with both harmonic index (p = 0.70, P = 0.03) and first maximum of the autocorrelation function (p = 0.89, P = 0.001). We conclude that (1) the tremor of workers exposed to Mn could be adequately described with only two variables; (2) a continuum of changes between tremor recorded in control subjects, in subjects exposed to Mn and in patients with PD was observed, with the MN group always found in between the control (CO) and the PD groups; (3) while blood Mn levels in workers were back at control levels at the time of the experiment, the effect of Mn on postural tremor was still detected. Thus our method has the potential to detect the effect of Mn on tremor with only two variables even after Mn level in the blood is back to normal values. (C)

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Bergeron RJ, Wiegand J, Weimar WR, Lindstrom TC, Fannin TL, Ratliff-Thompson K. 2004. Comparison of iron chelator efficacy in iron-overloaded beagle dogs and monkeys (Cebus apella). Comparative Medicine 54(6):664-672.
Abstract: Rodents and dogs are frequently used for preclinical toxicologic assessment of candidate iron chelators. Although the iron-clearing profile of a ligand often is known in rodents, and sometimes in primates, such information in dogs is rarely, if ever, available. Because of this, toxicity studies in dogs could be misleading; chelators that may otherwise be suitable for human clinical studies may be abandoned as being unacceptably toxic, simply because, unknown to the investigator, these drugs remove more iron in this species than would have been expected on the basis of iron clearance results in other species. This is a scenario that we encountered during toxicity trials of (S)-beta,beta-dimethylr4'-hydroxydesazadesmethyldesferrithiocin in dogs. Thus, we developed an iron-overloaded dog model in which it is possible to evaluate iron-clearing efficiencies of potential therapeutic ligands. Seven deferration agents have been screened in this model, and the results were compared with the iron-clearing efficiency of the same ligands in an iron-loaded Cebus apella monkey model. The data suggest that while the iron-clearing efficiencies of most of the drugs were similar between the two species, there can be profound differences. This is consistent with the idea that caution needs to be exercised when carrying out precliniclal toxicity evaluations of a chelator in dogs without first measuring the drug's iron-clearing efficiency in this species.

Berger MM, Jia XY, Legay V, Aymard M, Tilles JG, Lina B. 2004. Nutrition- and virus-induced stress represses the expression of manganese superoxide dismutase in vitro. Exp Biol Med 229(8):843-849.
Abstract: The relationship between oxidative stress and neuronal cell death has been suggested for many years. To understand the influence of oxidative stress on neuronal cell death, we investigated the influence of oxidative stress on DEV cells, a human glial cell line. Using enterovirus infection and/or malnutrition to induce oxidative stress, our results demonstrate that those stressors severely influence the antioxidant defense system in DEV cells. Although the expression of mitochondrial manganese superoxide dismutase (MnSOD) in DEV cells was significantly increased in acute infection with viral and nutritional stress, in persistent infection and nutritional stress, the expression of the MnSOD was drastically downregulated. We believe that this downregulation of MnSOD expression in the chronic stress model is due to repression of antioxidant defense. The downregulation of the MnSOD expression may lead to an increase of free-radical production and thus explain why the cells in the chronic stress model were more vulnerable to other oxidative stress influences. The vulnerability of DEV cells to additional stress factors resulted in progressive cell death, which may be analogous to the cell death in neurodegenerative diseases.

Berg D, Youdim M, Riederer P. 2004. Redox imbalance. Cell Tissue Res 318(1): 201-213.
Abstract: Substantial evidence implies that redox imbalance attributable to an overproduction of reactive oxygen species or reactive nitrogen species that overwhelm the protective defense mechanism of cells contributes to all forms of Parkinson's disease. Factors such as dopamine, neuromelanin, and transition metals may, under certain circumstances, contribute to the formation of oxygen species such as H2O2, superoxide radicals, and hydroxyl radicals and react with reactive nitrogen species such as nitric oxide or peroxinitrite. Mitochodrial dysfunction and excitotoxicity may be a cause and a result of oxidative stress. Consequences of this redox imbalance are lipid peroxidation, oxidation of proteins, DNA damage, and interference of reactive oxygen species with signal transduction pathways. These consequences become even more harmful when genetic variations impair the normal degradation of altered proteins. Therefore, therapeutic strategies must aim at reducing free-radical formation and scavenging

free-radicals.

Berg D, Hochstrasser H, Felletschin B, Deplazes J, Akbas N, Walter U. 2004. Genetic variations in brain iron metabolism in Parkinson's disease. Mov Disord 19:S199.

Ben Shachar D, Kahana N, Kampel V, Warshawsky A, Youdim MBH. 2004. Neuroprotection by a novel brain permeable iron chelator, VK-28, against 6-hydroxydopamine lession in rats. Neuropharmacology 46(2):254-263.
Abstract: Significant increase in iron occurs in the substantia nigra pars compacta of Parkinsonian subjects, and in 6-hydroxydopamine (6-OHDA) treated rats and monkeys. This increase in iron has been attributed to its release from ferritin and is associated with the generation of reactive oxygen species and the onset of oxidative stress-induced neurodegeneration. Several iron chelators with hydroxyquinoline backbone were synthesized and their ability to inhibit basal as well as iron-induced mitochondrial lipid peroxidation was examined. The neuroprotective potential of the brain permeable iron chelator, VK-28 (5-[4-(2-hydroxyethyl) piperazine-ylmethyl]-quinoline-8-ol), injected either intraventricularly (ICV) or intraperitoneally (IP), to 6-OHDA lesioned rats was investigated. VK-28 inhibited both basal and Fe/ascorbate induced mitochondrial membrane lipid peroxidation, with an IC50 (12.7 muM) value comparable to that of the prototype iron chelator, desferal, which does not cross the blood brain barrier. At an ICV pretreatment dose as low as 1 mug, VK-28 was able to completely protect against ICV 6-OHDA (250 pg) induced striatal dopaminergic lesion, as measured by dopamine (DA), dihydroxyphenyl acetic acid (DOPAC) and homovanilic acid (HVA) levels. IP injection of rats with VK-28 (1 and 5 mg/kg) daily for 10 and 7 days, respectively, demonstrated significant neuroprotection against lCV 6-OHDA at the higher dose, with 68% protection against loss of dopamine at 5mg/kg dosage of VK-28. The present study is the first to show neuroprotection with a brain permeable iron chelator. The latter can have implications for the treatment of Parkinson's disease and other neurodegenerative diseases (Alzheimer's disease, Friedreich ataxia, aceruloplasminemia, Hallervorden Spatz syndrome) where abnormal iron accumulation in the brain is thought to be associated with the degenerative processes. (C) 2003 Published by Elsevier Ltd.

Bates MN, Fawcett J, Garrett N, Cutress T, Kjellstrom T. 2004. Health effects of dental amalgam exposure: a retrospective cohort study. Int J Epidemiol 33 (4):894-902.
Abstract: Background Whether dental amalgam fillings (containing mercury) are hazardous is a long-standing issue, with few epidemiological investigations. Allegations have particularly involved nervous system disorders, such as multiple sclerosis, Alzheimer's disease, and chronic fatigue syndrome. This retrospective cohort study, the largest of its kind, contained people in the New Zealand Defence Force (NZDF) between 1977 and 1997. The NZDF has its own dental service, providing all personnel with regular and consistent treatment. Comprehensive treatment records are maintained and archived. Methods Yearly dental treatment histories, including amalgam filling placements, were compiled from individual records. To minimize amalgam exposure misclassification the cohort was restricted to people who, at NZDF entry, were aged <26 years and had all their posterior teeth. The cohort was linked with morbidity records. Data were analysed with a proportional hazards model, using a time-varying exposure unit of 100 amalgam surface-years. Results The final cohort contained 20 000 people, 84% males. Associations with medical diagnostic categories, particularly disorders of the nervous system and kidney, were examined. Of conditions allegedly associated with amalgam, multiple sclerosis had an adjusted hazard ratio (HR) of 1.24 (95% CI: 0.99, 1.53, P = 0.06), but there was no association with chronic fatigue syndrome (HR = 0.98, 95% CI: 0.94, 1.03), or kidney diseases. There were insufficient cases for investigation of Alzheimer's or Parkinson's diseases. Conclusions Results were generally reassuring, and provide only limited evidence of an association between amalgam and disease. Further follow-up of the cohort

will permit investigation of diseases more common in the elderly.

Bartzokis G, Tishler TA, Shin IS, Lu PH, Cummings JL. 2004. Brain Ferritin Iron as a Risk Factor for Age at Onset in Neurodegenerative DiseasesVolume 1012. p 224-236. Redox-Active Metals in Neurological Disorders: Annals of the New York Academy of Sciences.
Abstract: Tissue iron can promote oxidative damage. Brain iron increases with age and is abnormally elevated early in the disease process in several neurodegenerative disorders, including Alzheimer's disease (AD) and Parkinson's disease (PD). Higher iron levels in males may contribute to higher risk for younger-onset PD and recent studies have linked the presence of the hemo-chromatosis gene with a younger age at onset of AD. We examined whether age at onset of PD and AD was associated with increased brain ferritin iron. Ferritin iron can be measured with specificity in vivo with MRI utilizing the field-dependent relaxation rate increase (FDRI) method. FDRI was assessed in three basal ganglia regions (caudate, putamen, and globus pallidus) and frontal lobe white matter for younger- and older-onset male PD and AD patients and healthy controls. Significant increases in basal ganglia FDRI levels were observed in the younger-onset groups of both diseases compared to their respective control groups, but were absent in the older-onset patients. The results support the suggestion that elevated ferritin iron and its associated toxicity is a risk factor for age at onset of neurodegenerative diseases such as AD and PD. Clinical phenomena such as gender-associated risk of developing neurodegenerative diseases and the age at onset of such diseases may be associated with brain iron levels. In vivo MRI can measure and track brain ferritin iron levels and provides an opportunity to design therapeutic interventions that target high-risk populations early in the course of illness, possibly even before symptoms appear.

Bartzokis G. 2004. Quadratic trajectories of brain myelin content: unifying construct for neuropsychiatric disorders. Neurobiol Aging 25(1):49-62.
Abstract: Myelin plays an essential but largely underappreciated role in human brain structure and function. The central challenge raised by the six commentaries is whether the developmental model of age-related cognitive decline and Alzheimer's disease (AD) (Bartzokis, 2003, this issue) is applicable to a wider range of neurodegenerative and neuropsychiatric disorders. The model's premise that the trajectory of myelin development and breakdown is essential to our very uniqueness as a species, directly addresses this issue. In its widest perspective, the model primarily delineates a myelin hypothesis of human brain evolution and normal development and is "secondarily" useful in conceptualizing a wide range of age-related neuropsychiatric diseases. The unique vulnerabilities of oligodendrocytes and the highly protracted and extensive developmental process of human brain myelination delineated in the model are directly pertinent to many uniquely human brain functions and neuropsychiatric diseases including late-life neurodegenerative disorders. This lifelong perspective classifies AD as a disorder likely to arise in old age after a normal trajectory of myelin development. Genetic and environmental factors causing deviations in the myelination trajectory at any point in the lifespan will contribute to differences in the manifestations of later-life degenerative diseases and/or be detected in epidemiologic studies as risk factors or risk mitigators (age, cholesterol, iron, gender, education, brain, trauma, etc.). Ultimately, these perturbations of the myelination process could result in divergent-appearing disorders, such as frontotemporal dementia, dementia pugilistica, AD, dementia with Lewy bodies, and Parkinson's disease, that nonetheless have overlapping neuropathologic and/or clinical manifestations. Furthermore, the model's developmental perspective suggests that dysregulation in the uniquely vulnerable myelination process also contributes to highly prevalent early-life psychiatric disorders such as autism, attention deficit, schizophrenia, addiction, as well as their striking male predominance. Eventually, the dysregulated myelination associated with such disorders may have a direct and predictable impact on the appearance and manifestations of the later-life dementias. By increasing the scientific focus on the process of

myelination, this model may facilitate our understanding of the pathophysiology of multiple disorders and pathophysiologic, processes that cut across our current classification of diseases. Ultimately, the model provides a rational framework for the development of novel, myelin-centered treatments that may have widespread efficacy across multiple disease states and could potentially be used in delaying or even preventing some of the most devastating of human disorders. (C) 2003 Elsevier Inc. All rights reserved.

Barnham KJ, Masters CL, Bush AI. 2004. Neurodegenerative diseases and oxidative stress. Nature Reviews Drug Discovery 3(3):205-214.
Abstract: Oxidative stress has been implicated in the progression of Alzheimer's disease, Parkinson's disease and amyotrophic lateral sclerosis. Oxygen is vital for life but is also potentially dangerous, and a complex system of checks and balances exists for utilizing this essential element. Oxidative stress is the result of an imbalance in pro-oxidant/antioxidant homeostasis that leads to the generation of toxic reactive oxygen species. The systems in place to cope with the biochemistry of oxygen are complex, and many questions about the mechanisms of oxygen regulation remain unanswered. However, this same complexity provides a number of therapeutic targets, and different strategies, including novel metal - protein attenuating compounds, aimed at a variety of targets have shown promise in clinical studies.

Barhourni R, Faske J, Liu XH, Tjalkens RB. 2004. Manganese potentiates lipopolysaccharide-induced expression of NOS2 in C6 glioma cells through mitochondrial-dependent activation of nuclear factor kappaB. Molecular Brain Research 122(2):167-179.
Abstract: Neuronal injury in manganese neurotoxicity (manganism) is thought to involve activation of astroglial cells and subsequent overproduction of nitric oxide (NO) by inducible nitric oxide synthase (NOS2). Manganese (Mn) enhances the effects of proinflammatory cytokines on expression of NOS2 but the molecular basis for this effect has not been established. It was postulated in the present studies that Mn enhances expression of NOS2 through the cis-acting factor, nuclear factor kappaB (NF-kappaB). Exposure of C6 glioma cells to lipopopolysaccharide (LPS) resulted in increased expression of NOS2 and production of NO that was dramatically potentiated by Mn and was blocked through overexpression of mutant IkappaBalpha (S32/36A). LPS-induced DNA binding of p65/p50 was similarly enhanced by Mn and was decreased by mutant IkappaBalpha. Phosphorylation of IkappaBalpha was potentiated by Mn and LPS and was not blocked by U0126, a selective inhibitor of ERK1/2. Mn decreased mitochondrial membrane potential and increased matrix calcium, associated with a rise in intracellular reactive oxygen species (ROS) that was attenuated by the mitochondrial-specific antioxidant, MitoQ. Blocking mitochondrial ROS also attenuated the enhancing effect of Mn on LPS-induced phosphorylation of IkappaBalpha and expression of NOS2, suggesting a link between Mn-induced mitochondrial dysfunction and activation of NF-kappaB. Overexpression of a dominant-negative mutant of the NF-kappaB-interacting kinase (Nik) prevented enhancement of LPS-induced phosphorylation of IkappaBalpha by Mn. These data indicate that Mn augments LPS-induced expression of NOS2 in C6 cells by increasing mitochondrial ROS and activation of NF-kappaB. (C) 2004 Elsevier B.V. All rights reserved.

Atasoy HT, Nuyan O, Tunc T, Yorubulut M, Unal AE, Inan LE. 2004. T2-weighted MRI in Parkinson's disease; Substantia-nigra pars compacta hypointensity correlates with the clinical scores. Neurol India 52(3):332-337.
Abstract: Background: Iron accumulation in substantia nigra. pars compacta (SNpc) and related intensity and volumetric changes in patients with idiopathic Parkinson's disease (PD) has been reported previously. There are only a few studies evaluating the relation between neuroradiological findings and clinical scores, with contradictory results. Alms: In this study we aimed to measure the iron-rich brain areas of PD patients and healthy subjects with T2-weighted magnetic resonance

imaging (MRI) and to evaluate the relation between the clinical scores of PD patients and these imaging results. Methods and Materials: T2-weighted MRI findings were studied in 20 patients with PD and 16 healthy controls. The width of SNpc, putamen volume, and the intensity of the basal ganglia were measured. Unified Parkinson's Disease Rating Scale (UPDRS) was used for evaluating the clinical status. Statistical Analyses: Mann Whitney U test for group comparisons, Wilcoxon sign rank test for comparisons within the patient group, and Spearman's rank correlation coefficient for analyses of correlations were used. Results: Mean SNpc and dentate nucleus intensities were lower in PD patients than healthy subjects. Mean SNpc width and putamen volumes were lower in patients. Decrease in the intensity of mean SNpc correlated with high UPDRS and rigidity scores. Conclusion: The results of our study reflect the increase in iron accumulation and oxidative stress in the SNpc in Parkinson's disease. The decrease in the intensity of SNpc correlates with poor clinical scores.

Arnhold S, Semkova I, Andressen C, Lenartz D, Meissner G, Sturm V, Kochanek S, Addicks K, Schraermeyer U. 2004. Iris pigment epithelial cells: a possible cell source for the future treatment of neurodegenerative diseases. Exp Neurol 187(2):410-417.
Abstract: For the treatment of neurodegenerative disorders such as Parkinson's disease cell or gene therapeutical options are increasingly verified. For such approaches, neural stem cells or astrocytes are discussed as possible cell candidates. As also fetal retinal pigment epithelial cells have been successfully tested for such therapeutical options, we investigated the potential of iris pigment epithelial cells as an autologous source for future cell replacement therapies. Using the ELISA technique, we looked for the secretion of neurotrophic factors under basal and stimulated conditions by iris pigment epithelial cells (IPE) cells and compared them with the secretion of retinal pigment epithelial cells (RPE) cells. As iron plays a causative role in cell death during Parkinson's disease, the iron-binding capacity by IPE cells was investigated. Furthermore, we checked the integrative capacity of WE cells after transplantation into the striatum of adult rats. Our data reveal that IPE cells produce and secrete a variety of neurotrophic factors which can be stimulated after treatment with cytokines. Following transplantation, the cells can be easily detected by their pigmentation, survive for at least 8 weeks and as shown by electron microscopy integrate within the host tissue. Moreover, cells can be transduced with high efficiency using a third generation adenoviral vector, making them promising vehicles to locally deliver therapeutic proteins for the treatment of neurodegenerative diseases in a combined cell and gene therapeutical approach. (C) 2004 Elsevier Inc. All rights reserved.

Aoki I, Wu YJL, Silva AC, Lynch RM, Koretsky AP. 2004. In vivo detection of neuroarchitecture in the rodent brain using manganese-enhanced MRI. Neuroimage 22(3):1046-1059.
Abstract: Visualizing brain anatomy in vivo could provide insight into normal and pathophysiology. Here it is demonstrated that neuroarchitecture can be detected in the rodent brain using MRI after systemic MnCl2. Administration of MnCl2 leads to rapid T-1 enhancement in the choroid plexus and circumventricular organs, which spreads to the CSF space in ventricles and periventricular tissue. After 1 day, there was MRI enhancement throughout the brain with high intensity in the pituitary, olfactory bulb, cortex, basal forebrain, hippocampus, basal ganglia, hypothalamus, amygdala, and cerebellum. Contrast obtained enabled visualization of specific features of neuroarchitecture. The arrowhead structure of the dentate gyrus as well as the CA1 - CA3 region of the hippocampus and layers in cortex, cerebellum, as well as the olfactory bulb could be readily observed. Preliminary assignments of olfactory bulb layers, cortical layers in frontal and somatosensory cortex, and cerebellum were made. Systemic MnCl2 leads to MRI visualization of neuroarchitecture nondestructively. (C) 2004 Elsevier Inc. All rights reserved.

Andersen JK. 2004. Iron dysregulation and Parkinson's disease. Journal of

Alzheimers Disease 6(6):S47-S52.
Abstract: We have recently demonstrated that chelation of in vivo brain iron in a form which is not available to participate in oxidative events protects against a toxin-induced form of Parkinsonism in rodents, the well-established MPTP model [32]. These data strongly suggest that iron elevations observed in the Parkinsonian substantia nigra (SN), the brain region which undergoes selective neurodegeneration in the disease, are actively involved in subsequent neurodegenerative events. However the mechanism(s) by which iron levels become elevated in the Parkinsonian SN are still unclear. We hypothesize that increased oxidative stress associated with the disease may result in dysregulation of iron homeostasis in midbrain dopaminergic neurons via alterations in binding of iron regulatory proteins (IRPs). This would mechanistically explain the noted increase in cellular iron levels in the Parkinsonian SN which appear to contribute to subsequent neurodegeneration.

Anantharam V, Kitazawa M, Latchoumycandane C, Kanthasamy A, Kanthasamy AG. 2004. Blockade of Pkc Delta Proteolytic Activation by Loss of Function Mutants Rescues Mesencephalic Dopaminergic Neurons From Methylcyclopentadienyl Manganese Tricarbonyl (Mmt)-Induced Apoptotic Cell DeathVolume 1035. p 271-289. Protective Strategies for Neurodegenerative Diseases: Annals of the New York Academy of Sciences.
Abstract: The use of methytcyclopentadienyl manganese tricarbonyl (MMT) as a gasoline additive has raised health concerns and increased interest in understanding the neurotoxic effects of manganese. Chronic exposure to inorganic manganese causes Manganism, a neurological disorder somewhat similar to Parkinson's disease. However, the cellular mechanism by which MMT, an organic manganese compound, induces neurotoxicity in dopaminergic neuronal cells remains unclear. Therefore, we systematically investigated apoptotic cell-signaling events following exposure to 3-200 mu M MMT in mesencephalic dopaminergic neuronal (N27) cells. MMT treatment resulted in a time- and dose-dependent increase in reactive oxygen species generation and cell death in N27 cells. The cell death was preceded by sequential activation of mitochondrial-dependent proapoptotic events including cytochrome c release, caspase-3 activation, and DNA fragmentation, indicating that the mitochondrial-dependent apoptotic cascade primarily triggers MMT-induced apoptotic cell death. Importantly, MMT induced proteolytic cleavage of protein kinase C delta (PKC delta), resulting in persistently increased kinase activity. The proteolytic activation of PKC delta was suppressed by treatment with 100 mu M Z-VAD-FMK and 100 mu M Z-DEVD-FMK, suggesting that caspase-3 mediates the proteolytic activation of PKC delta. Pretreatment with 100 mu M Z-DEVD-FMK and 5 mu M rottlerin (a PKC delta inhibitor) also significantly attenuated MMT-induced DNA fragmentation. Furthermore, overexpression of either the kinase inactive dominant negative PKC delta(K376R) mutant or the caspase cleavage resistant PKC delta(D327A) mutant rescued N27 cells from MMT-induced DNA fragmentation. Collectively, these results demonstrate that the mitochondrial-dependent apoptotic cascade mediates apoptosis via proteolytic activation of PKC delta in MMT-induced dopaminergic degeneration and suggest that PKC delta may serve as an attractive therapeutic target in Parkinson-related neurological diseases.

Allkemper T, Schwindt W, Maintz D, Heindel W, Tombach B. 2004. Sensitivity of T2-weighted FSE sequences towards physiological iron depositions in normal brains at 1.5 and 3.0 T. Eur Radiol 14(6):1000-1004.
Abstract: To evaluate the sensitivity of T2-weighted fast spin-echo (FSE) sequences to physiological iron depositions in normal brains at MR imaging field strengths of 1.5 and 3.0 T. T2-weighted FSE sequences acquired at 1.5 and 3.0 T clinical imaging systems (Gyroscan Intera, Philips Medical Systems, Best, The Netherlands) were compared by means of MRI in phantoms (n=6) and healthy volunteers (n=10). Contrast-to-noise ratios (CNRs) of tubes doped with iron oxides at different concentrations and of brain areas with physiological iron depositions (nucleus ruber, substantia nigra, globus pallidus) were calculated for either field strength. Apparent

susceptibility effects of iron-containing brain structures were qualitatively analyzed by comparing the degree of visible hypointensity by a score system at either field strength. The mean CNR of iron oxide tubes and iron-containing brain areas was significantly decreased at 3.0 T. Qualitative analysis confirmed these measurements. Detection and diagnosis of brain disorders with altered iron content such as neurodegenerative parkinsonian disorders (NPD) or intracerebral hemorrhage should benefit from the increased sensitivity of T2-weighted FSE sequences to susceptibility effects at 3.0 T.

Akundi RS, Macho A, Munoz E, Lieb K, Bringmann G, Clement HW, Hull M, Fiebich BL. 2004 . 1-trichloromethyl-1,2,3,4-tetrahydro-beta-carboline-induced apoptosis in the human neuroblastoma cell line SK-N-SH. J Neurochem 91 (2):263-273.
Abstract: Trichloroethylene, a common industrial solvent and a metabolic precursor of chloral hydrate, occurs widely in the environment. Chloral hydrate, which is also used as a hypnotic, has been found to condense spontaneously with tryptamine, in vivo, to give rise to a highly unpolar 1-trichloromethyl-1,2,3,4-tetrahydro-beta-carboline (TaClo) that has a structural analogy to the dopaminergic neurotoxin N-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP). Earlier studies have revealed the relative permeability of the molecule through the blood-brain barrier and its ability to induce Parkinson-like symptoms in rats. In this study, we report that TaClo induces an apoptotic pathway in the human neuroblastoma cell line, SK-N-SH, involving the translocation of mitochondrial cytochrome c to the cytosol and activation of caspase 3. TaClo-induced apoptosis shows considerable differences from that mediated by other Parkinson-inducing agents such as MPTP, rotenone and manganese. Although it is not clear if the clinically administered dosage of chloral hydrate or the relatively high environmental levels of trichloroethylene could lead to an onset of Parkinson's disease, the spontaneous in vivo formation of TaClo and its pro-apoptotic properties, as shown in this report, should be considered.

Agostinelli E, Arancia G, Dalla Vedova L, Belli F, Marra M, Salvi M, Toninello A. 2004 . The biological functions of polyamine oxidation products by amine oxidases: Perspectives of clinical applications. Amino Acids 27(3-4): 347-358.
Abstract: The polyamines spermine, spermidine and putrescine are ubiquitous cell components. If they accumulate excessively within the cells, due either to very high extracellular concentrations or to deregulation of the systems which control polyamine homeostasis, they can induce toxic effects. These molecules are substrates of a class of enzymes that includes monoamine oxidases, diamine oxidases, polyamine oxidases and copper containing amine oxidases. Polyamine concentrations are high in growing tissues such as tumors. Amine oxidases are important because they contribute to regulate levels of mono- and polyamines. These enzymes catalyze the oxidative deamination of biogenic amines and polyamines to generate the reaction products H2O2 and aldehyde(s) that are able to induce cell death in several cultured human tumor cell lines. H2O2 generated by the oxidation reaction is able to cross the inner membrane of mitochondria and directly interact with endogenous molecules and structures, inducing an intense oxidative stress. Since amine oxidases are involved in many crucial physiopathological processes, investigations on their involvement in human diseases offer great opportunities to enter novel classes of therapeutic agents.

Cookson MR, Lockhart PJ, McLendon C, O'Farrell C, Schlossmacher M, Farrer MJ. 2003 Nov 15. RING finger 1 mutations in Parkin produce altered localization of the protein. Hum Mol Genet 12(22):2957-65.
Abstract: The Parkin gene (PRKN) encodes an E3 protein-ubiquitin ligase for which loss of function is associated with autosomal-recessive juvenile (<20 years) and early-onset Parkinsonism (<45 years). Although detailed pathological reports are scarce, brains from patients with homozygous exonic deletions demonstrate neuronal loss in the substantia nigra, albeit

without the Lewy body pathology characteristic of idiopathic Parkinson's disease. However, there are rare descriptions of more florid pathology, including Lewy bodies and tau positive astrocytes in individuals with compound heterozygous mutations. In the present study we examined whether PRKN point mutations, leading to amino acid substitutions, may alter the cellular distribution of the protein produced. Wild-type Parkin was homogeneously distributed throughout the cytoplasm with a small amount of protein in the nucleus after transfection into human embryonic kidney cells. Mutant isoforms with A82E, G328E and C431F amino acid substitutions were also normally distributed. However, two mutant isoforms, R256C and R275W, within RING finger 1 of the Parkin protein (238-293 amino acids), produced an unusual distribution of the protein, with large cytoplasmic and nuclear inclusions. We have replicated this observation in primary cultured neurons and demonstrate, by the accumulation/co-localization of cytoskeletal protein vimentin, that the inclusion bodies are aggresomes, a cellular response to misfolded protein.

Jiang H, Qian ZM, Xie JX. 2003 Oct 25. [Increased DMT1 expression and iron content in MPTP-treated C57BL/6 mice]. Sheng Li Xue Bao 55(5):571-6.
Abstract: Iron plays a key role in Parkinson s disease (PD). To illustrate the mechanism underlying the increase of iron in substantia nigra (SN) in PD, changes of the expression of divalent metal transporter 1 (DMT1) and iron content were examined in SN in 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) treated mice using immunohistochemistry and histochemistry respectively. Following MPTP treatment for 3 d, elevated iron staining was found in SN. A further increase in iron content was observed after 7 d. In these lesioned animals, tyrosine hydroxylase-immunoreactive DA neurons exhibited a decrease in number and morphological changes as well. There were two isoforms of DMT1 expressed in SN of mice. After MPTP treatment, the expression of DMT1 without IRE form increased in either group, whereas DMT1 with IRE form increased only after 7 d of MPTP treatment. These observations suggest that DMT1 is possibly involved in the process of iron accumulation in SN of MPTP-treated mice, which might be responsible for the subsequent death of DA neurons.

Hebert G, Arsaut J, Dantzer R, Demotes-Mainard J. 2003 Oct 9. Time-course of the expression of inflammatory cytokines and matrix metalloproteinases in the striatum and mesencephalon of mice injected with 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine, a dopaminergic neurotoxin. Neurosci Lett 349(3):191-5.
Abstract: Injection of 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) in mice results in a retrograde nigrostriatal dopaminergic pathway denervation and subsequent tissue reorganization. Since the role of inflammatory mediators after MPTP remains unclear, proinflammatory cytokine and matrix metalloproteinase (MMP) expression were evaluated by comparative RT-PCR during denervation and tissue reorganization following a single-dose of MPTP (40 mg/kg, s.c.) in young (8-week-old) mice. The time-course of denervation/reorganization was assessed through [(3)H]GBR-12935 binding on dopamine transporter and tyrosine hydroxylase immunohistochemistry. In the striatum, TNF-alpha, IL-1alpha, IL-1beta, IL-6 and MMP-9 mRNA expression peaked on day 1. In the ventral mesencephalon, cytokines (TNF-alpha, IL-1alpha, IL-1beta) and MMP-9 mRNA expression peaked on day 3. During tissue reorganization (day 6 through 16), the only change observed in the striatum consisted of IL-1alpha mRNA and protein overexpression together with MMP-2 downregulation. Whereas the early expression of proinflammatory cytokines and MMP might participate in the retrograde nigrostriatal denervation, the late component of IL-1alpha expression suggests a possible role for this cytokine in the subsequent striatal reorganization.

Junxia X, Hong J, Wenfang C, Ming Q. 2003 Aug. Dopamine release rather than content in the caudate putamen is associated with behavioral changes in the iron rat model of Parkinson's disease. Exp Neurol 182(2):483-9.
Abstract: The effects of intranigral iron injection on dopamine (DA) release

and content in the caudate putamen (CPu) and their relationship to DA-related behavioral response were investigated in rats. Different concentrations of FeCl(3) (10, 20, and 40 microg) and saline were injected separately into the left substantia nigra. In some experiments, rats were pretreated with desferrioxamine or saline before iron injection. After 3 weeks, changes in behavioral response, DA release, and DA content in the CPu were determined. In all iron injection groups (10, 20, and 40 microg), DA content in the lesioned side of the brain was significantly decreased, showing a significant linear correlation (R(2) = 0.981, P = 0.01), and DA turnover ratio significantly increased (both P = 0.01, 0.01 and 0.001 vs unlesioned sides, respectively). However, injection dosages of 10 or 20 microg of iron did not lead to significant changes in DA release in the CPu or in behavioral response. At the 40-microg dosage, it was found that DA release in the lesioned side and rearing activity both were significantly reduced (all P = 0.01 vs unlesioned side or control) and apomorphine-induced rotation was observed. Pretreatment with desferrioxamine significantly inhibited the effect of iron on DA release and content. These results demonstrate that iron injection can damage dopaminergic neurons and suggest that DA release, rather than DA content, in the CPu is associated with DA-related behavioral changes in this PD model.

Lucchini R, Benedetti L, Borghesi S, Garattini S, Parrinello G, Alessio L. 2003 Jul-Sep. [Exposure to neurotoxic metals and prevalence of parkinsonian syndrome in the area of Brescia]. G Ital Med Lav Ergon 25 Suppl(3):88-9. Abstract: The prevalence of parkinsonian syndromes was studied in the province of Brescia (Northern Italy), in order to verify its possible increase in the surroundings of ferroalloy plants located in a valley of the pre-Alps. A case-list of subjects affected by these disturbances was identified using four different sources of information: a) registers from local medical clinics; b) admission charts from local hospitals; c) consumption of levodopa; d) NHS list of exemption from prescription payment, due to the illness. Exploratory data show a frequency of parkinsonian disturbances among the residents in the surroundings of the ferroalloy plants and downwind (crude prevalence = 358/100,000 population, standardized for age and sex = 438) significantly higher (s.m.r, = 1.58; C.I. = 1.41-1.76) than the entire Province (crude prevalence 246/100,000). This preliminary result could indicate the interaction of prolonged environmental exposure to heavy metals, such as manganese, and genetic factors, potentially relevant in this mountain population.

Garcia de Yebenes J. 2003 Jul-Aug. [Zoofilia as late complication of Parkinson's disease. Circe drugs and Mercury antidotes]. Neurologia 18(6):351.

Yomono H, Kurisaki H, Murayama S, Hebisawa A, Miyajima H, Takahashi Y. 2003 Jul. [An autopsy case of multiple system atrophy with a heteroallelic ceruloplasmin gene mutation]. Rinsho Shinkeigaku 43(7):398-402. Abstract: We reported a 69-year-old woman with multiple system atrophy (MSA), who had a heteroallelic missense mutation (G1874A, Gly-->Glu) in the exon 11 of the ceruloplasmin (Cp) gene. At the age of 64, she began to complain of progressive gait disturbance, which was resistant to anti-Parkinsonian drug treatment. Neurological examination revealed parkinsonism such as rigidity, akinesia, mild tremor and postural instability, accompanying saccadic eye movement, dysarthria, dysphagia, orthostatic hypotension and bladder disturbance. She showed neither cerebellar signs nor dementia. Serum Cp and copper concentrations were 13-18 mg/dl and 38-56 micrograms/dl, respectively, which were decreased to about a half of normal values. Brain MRIs revealed high intensity areas in the bilateral putamens in the T2-weighted image, and mild pontine base atrophy. She died of respiratory failure due to laryngeal paresis after five years from the onset. Neuropathological examination revealed brown-colored putamens, where there was severe neuronal cell loss with gliosis. Though atrophy of the pontine base was mild, transverse myelinated fibers were pale in Kluver-Barrera stain. There were Purkinje cell loss of moderate degree and appearance of torpedos in the cerebellum. Both silver staining and immunohistochemical staining to alpha-synuclein

showed glial cytoplasmic inclusions, which were found predominantly in the putamens. These clinical features and neuropathological findings were compatible with multiple system atrophy (MSA). Iron staining of the brain revealed iron deposition in the putamens and the substantia nigra, but not in the pontine base nor in the cerebellum. Furthermore, we failed to reveal it in both the liver and the pancreas as well as the thalamus and the caudate nucleus, which were common sites of iron deposition in the previous cases of Cp gene mutation. We have already reported three other MSA cases with a- or hypo-ceruloplasminemia with similar clinical and pathological features to this case. One of them, in which gene analysis was also available, did not have any mutations in its Cp gene. Therefore, the gene mutation of this case may not be a direct cause to MSA, but the fact that the most cases of MSA with hypoceruloplasminemia showed striatonigral degeneration (SND) type implies some relationship between hypoceruloplasminemia and SND.

Kaiser J. 2003 May 9. State Court to rule on manganese fume claims. Science 300(5621):927.

Youdim MB, Amit T, Bar-Am O, Weinstock M, Yogev-Falach M. 2003 May. Amyloid processing and signal transduction properties of antiparkinson-antialzheimer neuroprotective drugs rasagiline and TV3326. Ann N Y Acad Sci 993:378-86; discussion 387-93.
Abstract: Two novel neuroprotective cholinesterase (ChE) inhibitors, TV3326 and TV3279 [(N-propargyl-(3R) and (3S) aminoindan-5-yl)-ethyl methyl carbamate], respectively were derived from rasagiline, for the treatment of Alzheimer's disease (AD). TV3326 also inhibits monoamine oxidase (MAO)-A and B, while its S-isomer, TV3279, lacks MAO-inhibitory activity. The actions of these drugs in the regulation of the amyloid precursor protein (APP) processing using rat PC12 and human SH-SY5Y neuroblastoma cells were examined. Both isomers stimulated the release of the non-amyloidogenic alpha-secretase form of soluble APP (sAPPalpha) from these cell lines. The increases in sAPPalpha, induced by TV3326 and TV3279, were dose-dependent (0.1-100 micro M) and blocked by the hydroxamic acid-based metalloprotease inhibitor, Ro31-9790, suggesting mediation via alpha-secretase activity. Using several signal transduction inhibitors, the involvement of protein kinase C (PKC), mitogen-activated protein (MAP) kinase, and tyrosine kinase-dependent pathways in the enhancement of sAPPalpha release by TV3326 and TV3279 was identified. In addition, both drugs directly induced the phosphorylation of p44 and p42 MAP kinase, which was abolished by the specific inhibitors of MAP kinase activation, PD98059 and U0126. These data suggest a novel pharmacological mechanism, whereby these ChE inhibitors regulate the secretary processes of APP via activation of the MAP kinase pathway.

Cole GM. 2003 Mar 27. Ironic fate: can a banned drug control metal heavies in neurodegenerative diseases? Neuron 37(6):889-90.
Abstract: In this issue of Neuron, Kaur et al. demonstrate that iron chelation by ferritin transgene or the metal chelator clioquinol prevent oxidative damage and MPTP toxicity in mice. This raises the issue of specific iron chelators or clioquinol for control of oxidative damage in Parkinson's, Alzheimer's, and other neurodegenerative diseases, but not without safety concerns.

Uc EY, Rodnitzky RL. 2003 Mar. Childhood dystonia. Semin Pediatr Neurol 10(1): 52-61.
Abstract: Childhood dystonias are a heterogeneous group of disorders with strong inherited basis. This review describes the clinical characteristics, classification, genetic basis, pathophysiology, biochemistry, pathology, and treatment of dystonias, including the primary dystonias, the dystonia-plus syndromes, secondary dystonias, and heredodegenerative disorders. Conditions discussed in detail include idiopathic torsion dystonia, dopa-responsive dystonia, Wilson's disease, myoclonus dystonia, rapid-onset dystonia parkinsonism, neurodegeneration with brain iron accumulation (Hallervorden-Spatz syndrome), mitochondrial dystonias, Niemann-Pick

type C, and neuroacanthocytosis.

Wang J, Jiang H, Xie JX. 2003 Jan. [The relationship between iron metabolism in central nervous system and Parkinson's disease]. Sheng Li Ke Xue Jin Zhan 34(1):67-70.

Zhang SR, Zhou ZC, Fu JL. 2003. Effect of manganese chloride exposure on liver and brain mitochondria function in rats. Environ Res 93(2):149-157.
Abstract: Manganese (Mn) is an essential trace element found in many enzymes. As is the case for many essential trace elements, excessive Mn is toxic. Individuals suffering from manganese toxicity exhibit several symptoms, which are similar to those frequently observed in cases of Parkinson's disease. In this investigation, we studied the effect of manganese chloride (7.5, 15.0, and 30.0 mg/kg body weight) on mitochondrial function and attempted to ascertain the mechanism of manganese-induced mitochondrial dysfunction. The production of reactive oxygen species in mitochondria of rat liver and brain was assayed using 2,7'-dichlorofluorescin diacetate, and the activities of respiratory chain enzymes were examined spectrophotometrically. Monoamine oxidase (MAO) activity was assayed by measuring reduction of benzylamine. Manganese and calcium content in mitochondria were determined by atomic absorption spectrophotometry. These results indicate that manganese chloride (MnCl2) can decrease MAO activity and inhibit the respiratory chain. Manganese can accumulate in mitochondria and inhibit efflux of calcium. There is a significant inverse correlation between the amount of superoxide radicals and the specific activities of the mitochondria enzymes. Mitochondrial function was significantly affected in both males and females. (C) 2003 Elsevier Inc. All rights reserved.

Zhang J, Fitsanakis VA, Gu GY, Jing DQ, Ao MF, Amarnath V, Montine TJ. 2003. Manganese ethylene-bis-dithiocarbamate and selective dopaminergic neurodegeneration in rat: a link through mitochondrial dysfunction. J Neurochem 84(2):336-346.
Abstract: Manganese ethylene-bis-dithiocarbamate (Mn-EBDC) is the major active element of manab, a pesticide linked to parkinsonism in certain individuals upon chronic exposure. Additionally, it has been shown to produce dopaminergic neurodegeneration in mice systemically coexposed to another pesticide, 1,1'-dimathyl-4,4'-bipyridinium (paraquat). Here, we described a rat model in which selective dopaminergic neurodegeneration was produced by delivering Mn-EBDC directly to the lateral ventricles. After establishing this model, we tested whether Mn-EBDC provoked dopamine efflux in the striatum, a well-known phenomenon produced by the mitochondrial inhibitor 1-methyl-4-phenylpyridinium (MPP), the active metabolite of 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) that causes parkinsonism in humans, as well as in some animals. Finally, we investigated whether Mn-EBDG directly inhibited mitochondrial function in vitro using isolated brain mitochondria. Our data demonstrated that Mn-EBDG induced extensive striatal dopamine efflux that was comparable with that induced by MPP+, and that Mn-EBDC preferentially inhibited mitochondrial complex Ill. As mitochondrial dysfunction is pivotal in the pathogenesis of Parkinsons disease (PD), our results support the proposal that exposure to pesticides such as maneb, or other naturally occurring compounds that inhibit mitochondrial function, may contribute to PD development.

Zecca L, Zucca FA, Wilms H, Sulzer D. 2003. Neuromelanin of the substantia nigra: a neuronal black hole with protective and toxic characteristics. Trends Neurosci 26(11):578-580.
Abstract: Neuromelanin accumulates in dopaminergic neurons during normal aging, and in Parkinson's disease, neurons with this pigment are those that selectively degenerate. Intraneuronal neuromelanin could play a protective role during its synthesis by preventing the toxic accumulation of cytosolic catechol derivatives and, in addition, by its ability to scavenge reactive metals, pesticides and other toxins to form stable adducts. However, dying neurons in Parkinson's disease that release neuromelanin

might induce a vicious cycle of chronic neuroinflammation and neuronal loss.

Zecca L, Zucca FA, Costi P, Tampellini D, Gatti A, Gerlach M, Riederer P, Fariello RG, Ito S, Gallorini M, Sulzer D. 2003. The neuromelanin of human substantia nigra: structure, synthesis and molecular behaviour. Journal of Neural Transmission-Supplement (65):145-155.
Abstract: The pigmented neurons of the substantia nigra (SN) are typically lost in Parkinson's disease: however the possible relationship between neuronal vulnerability and the presence of neuromelanin (NM) has not been elucidated. Early histological studies revealed the presence of increasing amounts of NM in the SN with aging in higher mammals, showed that NM granules are surrounded by membrane, and comparatively evaluated the pigmentation of SN in different animal species. Histochemical studies showed the association of NM with lipofuscins. However, systematic investigations of NM structure, synthesis and molecular interactions have been undertaken only during the last decade. In these latter studies, NM was identified as a genuine melanin with a strong chelating ability for iron and affinity for compounds such as lipids, pesticides, and MPP+. The affinity of NM for a variety of inorganic and organic toxins is consistent with a postulated protective function for NM. Moreover, the neuronal accumulation of NM during aging, and the link between its synthesis and high cytosolic concentration of catechols suggests a protective role. However, its putative neuroprotective effects could be quenched in conditions of toxin overload.

Zatta P, Lucchini R, Van Rensburg SJ, Taylor A. 2003. The role of metals in neurodegenerative processes: aluminum, manganese, and zinc. Brain Res Bull 62(1):15-28.
Abstract: Until the last decade, little attention was given by the neuroscience community to the neurometabolism of metals. However, the neurobiology of heavy metals is now receiving growing interest, since it has been linked to major neurodegenerative diseases. In the present review some metals that could possibly be involved in neurodegeneration are discussed. Two of them, manganese and zinc, are essential metals while aluminum is non-essential. Aluminum has long been known as a neurotoxic agent. It is an etiopathogenic factor in diseases related to long-term dialysis treatment, and it has been controversially invoked as an aggravating factor or cofactor in Alzheimer's disease as well as in other neurodegenerative diseases. Manganese exposure can play an important role in causing Parkinsonian disturbances, possibly enhancing physiological aging of the brain in conjunction with genetic predisposition. An increased environmental burden of manganese may have deleterious effects on more sensitive subgroups of the population, with sub-threshold neurodegeneration in the basal ganglia, generating a pre-Parkinsonian condition. In the case of zinc, there has as yet been no evidence that it is involved in the etiology of neurodegenerative diseases in humans. Zinc is redox-inactive and, as a result of efficient homeostatic control, does not accumulate in excess. However, adverse symptoms in humans are observed on inhalation of zinc fumes, or accidental ingestion of unusually large amounts of zinc. Also, high concentrations of zinc have been found to kill bacteria, viruses, and cultured cells. Some of the possible mechanisms for cell death are reviewed. (C) 2003 Elsevier Inc. All rights reserved.

Zambrzycka A, Cakala M, Kaminska M. 2003. Transition metal ions significantly decrease phospholipase C activity degrading phosphatidylinositol-4,5-bisphosphate in the brain cortex. Pol J Pharmacol 55(5):915-917.
Abstract: Highly reactive transition metals, such as copper and iron play an obligatory role in generating of reactive oxygen species (ROS). Many neurodegenerative diseases including Alzheimer's disease (AD) and Parkinson's disease (PD) show increased accumulation of these metals. Phosphoinositide metabolism is altered in neurodegenerative diseases. In the present study, we examined the effect of CuSO4 and FeCl2 on phospholipase C (PLC) activity degrading phosphatidylinositol-4,5-bisphosphate (PIP2) and phosphatidylinositol (PI) in synaptic plasma

membranes (SPM) from the rat brain cortex. We report that 25 muM CuSO4 and FeCl2 decreased PIP2-PLC activity by 60% and 75%, respectively. However, both compounds had no effect on PI-PLC activity. These data indicated that exclusively PIP2-PLC is sensitive to transition metal ions. We suggest that chelators of these metals may protect brain against alteration of phosphoinositide metabolism and might be beneficial in the treatment of neurodegenerative diseases.

Youdim MBH, Mandel S, Maor G, Levites Y. 2003. Iron chelators-radical scavengers 3,3-epigallocatechin-3-gallate (EGCG) from tea extract and apomorphine attenuate neuronal cell death in 6-hydroxydopamine and MPTP models of Parkinson's Disease: Possible gene targets employing CDNA microarray. Biometals 16(1):228.

Youdim MBH. 2003. What have we learnt from CDNA microarray gene expression studies about the role of iron in MPTP induced neurodegeneration and Parkinson's disease? Journal of Neural Transmission-Supplement (65): 73-88.
Abstract: There have been numerous hypotheses concerning the etiology and mechanism of dorsal raphe dopaminergic neurodegeneration in Parkinson's disease and its animal models, MPTP (N-methyl-4-phenyl-1,2,3,6-tetrahydropyridine) and 6-hydroxydopamine. The advent of cDNA microarray gene expression where expression of thousands of genes can be globally assessed has indicated that mechanism of neurodegeneration by MPTP is a complex cascade of vicious circles. One of these is the alteration of genes associated with iron metabolism, a transitional metal closely associated with inducing the formation of reactive oxygen species and inducing oxidative stress. cDNA gene expression analyses support the established hypothesis of oxidative induced neurodegeneration involving iron deposition in substantia nigra pars compacta (SNPC) parkinsonian brains. The regulation of cellular iron metabolism has been further enhanced by the recent discovery of two iron regulatory proteins, IRP1 and IRP2 which control the level of iron with in the cell. When the cellular level of iron increases IRP2 is degraded by ubiquitination and no further iron accumulates. The reverse occurs when the level of iron is low within the cell. Knock-out IRP1 and IRP2 mice have shown that in latter mice brain iron accumulation precedes the neurodegeneration, ataxia and bradykinesia observed in these animals. Indeed MPTP treatment, which results in iron accumulation in SNCP, abolishes IRP2 with the concomitant increase in alpha-synuclein. Iron chelators such as R-apomorphine and EGCG, which protect against MPTP neurotoxicity, prevent the loss of IRP2 and the increase in alpha-synuclein. The presence of iron together with alpha-synuclein in SNPC may be detrimental for dopaminergic neurons. Since, iron has been shown to cause aggregation of alpha-synuclein to a neurotoxic agent. The use of iron chelators penetrating the blood brain barrier as neuroprotective drugs has been envisaged.

Youdim MB. 2003. What have we learnt from CDNA microarray gene expression studies about the role of iron in MPTP induced neurodegeneration and Parkinson's disease? J Neural Transm Suppl (65):73-88.
Abstract: There have been numerous hypotheses concerning the etiology and mechanism of dorsal raphe dopaminergic neurodegeneration in Parkinson's disease and its animal models, MPTP (N-methyl-4-phenyl-1,2,3,6-tetrahydropyridine) and 6-hydroxydopamine. The advent of cDNA microarray gene expression where expression of thousands of genes can be globally assessed has indicated that mechanism of neurodegeneration by MPTP is a complex cascade of vicious circles. One of these is the alteration of genes associated with iron metabolism, a transitional metal closely associated with inducing the formation of reactive oxygen species and inducing oxidative stress. cDNA gene expression analyses support the established hypothesis of oxidative induced neurodegeneration involving iron deposition in substantia nigra pars compacta (SNPC) parkinsonian brains. The regulation of cellular iron metabolism has been further enhanced by the recent discovery of two iron

regulatory proteins, IRP1 and IRP2 which control the level of iron with in the cell. When the cellular level of iron increases IRP2 is degraded by ubiquitination and no further iron accumulates. The reverse occurs when the level of iron is low within the cell. Knock-out IRP1 and IRP2 mice have shown that in latter mice brain iron accumulation precedes the neurodegeneration, ataxia and bradykinesia observed in these animals. Indeed MPTP treatment, which results in iron accumulation in SNCP, abolishes IRP2 with the concomitant increase in alpha-synuclein. Iron chelators such as R-apomorphine and EGCG, which protect against MPTP neurotoxicity, prevent the loss of IRP2 and the increase in alpha-synuclein. The presence of iron together with alpha-synuclein in SNPC may be detrimental for dopaminergic neurons. Since, iron has been shown to cause aggregation of alpha-synuclein to a neurotoxic agent. The use of iron chelators penetrating the blood brain barrier as neuroprotective drugs has been envisaged.

Yoshikawa K, Matsumoto M, Hamanaka M, Nakagawa M. 2003. A case of manganese induced parkinsonism in hereditary haemorrhagic telangiectasia. J Neurol Neurosurg Psychiatry 74(9):1312-1314.
Abstract: A 44 year old right handed woman complained of difficulty in moving. She and her relatives had skin telangiectasia or recurrent epistaxis. On neurological examination, she had a mask-like facies and bradykinesia in both extremities. Laboratory examinations showed iron deficiency anaemia and mild liver dysfunction with raised serum manganese. On T1 weighted cranial magnetic resonance imaging there were hyperintense areas in the globus pallidus bilaterally, suggesting manganese deposition. Abdominal angiography confirmed multiple portal-systemic shunts in the liver, and a needle biopsy of the liver showed diffuse dilatation of the sinusoids with fatty change. Levodopa did not improve the bradykinesia. This appears to be a case of hereditary haemorrhagic telangiectasia with manganese induced parkinsonism, which may be a new type of neurological disorder in such patients.

Yoshida S, Ide-Ektessabi A, Fujisawa S. 2003. Application of synchrotron radiation in neuromicrobiology: role of iron in Parkinson's disease. Structural Chemistry 14(1):85-95.
Abstract: In order to understand the role of iron (Fe) in the oxidative stress underlying the pathogenesis of Parkinson's disease (PD) and parkinsonism dementia complex (PDC), we investigate distributions and chemical states of Fe within a single neuron of the two disease cases, using synchrotron radiation (SR) micro beam. In the X-ray fluorescence (XRF) spectroscopic study, an excessive accumulation of Fe can be seen in the melanized neurons and free-neuromelanin (MN) aggregates in the substantia nigra tissue of both PD and PDC midbrains. X-ray absorption near-edge structure (XANES) analyses of PD revealed that the chemical state of Fe in the melanized neurons and free-MN aggregates shifted toward Fe3+ with a pre-edge peak at Fe K-edge due to a 1s --> 3d transition, indicating a breaking of the inversion symmetry around the Fe site. In PDC, however, the melanized neurons and free-MN aggregates showed mixed states of Fe2+ and Fe3+ without any pre-edge peak in the spectra. This tendency was also observed in the control case. These results suggest that the changes in distributions and chemical states of Fe may endogenously play a crucial role in the oxidative damage of the melanized neurons in PD, but through a different mechanism other than PDC.

Yogev-Falach M, Amit T, Bar-Am O, Youdim MBH . 2003. The importance of propargylamine moiety in the anti-Parkinson drug rasagiline and its derivatives for MAPK-dependent amyloid precursor protein processing. FASEB J 17(13).
Abstract: Rasagiline [N-propargyl-(1R)-aminoindan] a highly potent selective irreversible monoamine oxidase (MAO)-B inhibitor exerts neuroprotective and antiapoptotic effects against a variety of insults in cell cultures and in vivo and has finished its phase III clinical trials for Parkinson's disease. In the present study, we show that rasagiline (1 and 10 muM) significantly protected rat PC12 cells against beta-amyloid (Abeta

(1-42)) toxicity. In addition, rasagiline significantly increased (approximately threefold) the secretion of the nonamyloidogenic soluble form of the amyloid precursor protein (sAPPalpha) from SH-SY5Y neuroblastoma and PC12 cells. The increase of sAPPa. was dose-dependent and was blocked by the hydroxamic acid-based metalloprotease inhibitor Ro31-9790 (100 muM), suggesting that the effect is mediated via alpha-secretase activity. Rasagiline-induced sAPPa. release was significantly reduced by the inhibitors of protein kinase C (PKC), GF109203X, and ERK mitogen-activated protein kinase (MAPK) PD98059. Moreover, rasagiline dose dependently (0.1-10 muM) increased the phosphorylation of p44 and p42 MAPK, which was abolished by PD98059 (30 muM) and GF109203X (2.5 muM). By comparing the actions of rasagiline with those of its S-isomer TVP1022, which is not an MAO inhibitor, we have been able to demonstrate that MAO-B inhibition is not a prerequisite for either sAPPalpha-induced release or ERK phosphorylation. In addition, structure-activity relationship among rasagiline-related compounds suggests the crucial role of the propargyl moiety in these molecules, because propargylamine itself significantly induced the secretion of sAPPa. and increased MAPK phosphorylation with similar potency to that of rasagiline and its derivatives.

Yen JH, Tsai WC, Lin CH, Ou TT, Hu CJ, Liu HW. 2003. Manganese superoxide dismutase gene polymorphisms in psoriatic arthritis. Dis Markers 19(6): 263-265.
Abstract: The purpose of the present study is to investigate the role of manganese superoxide dismutase (MnSOD) gene polymorphisms in the susceptibility to psoriatic arthritis. MnSOD gene polymorphisms were determined by polymerase chain reaction/restriction fragment length polymorphisms method in fifty-two patients with psoriatic arthritis and 90 healthy controls. The genotype frequency of MnSOD 1183C/T was significantly higher in patients with psoriatic arthritis than in controls. In contrast, the frequency of MnSOD 1183T/T was significantly decreased in patients with psoriatic arthritis. The phenotype frequency of MnSOD 1183C was significantly increased in patients with psoriatics arthritis in comparison to healthy controls. Therefore, MnSOD 1183C polymorphisms may be a precipitating factor for the development of psoriatic arthritis.

Yen JH, Chen CJ, Tsai WC, Lin CH, Ou TT, Hu CJ, Liu HW. 2003. Manganese superoxide dismutase and cytochrome P450 1A1 genes polymorphisms in rheumatoid arthritis in Taiwan. Hum Immunol 64(3):366-373.
Abstract: To investigate the role of manganese superoxide dismutase (MnSOD) and cytochrome P450 1A1 (CYP1A1) gene polymorphisms in the pathogenesis of rheumatoid arthritis (RA) in Taiwan, MnSOD and CYP1A1 genes polymorphisms were determined by he polymerase chain reaction/ restriction fragment length polymorphism method in 112 patients with RA and 96 controls. There were no significant differences in the genotype, allele, and phenotype frequencies of MnSOD Ala-9Val (C1183T) polymorphisms between patients with RA and controls. The polymorphism of MnSOD 5777T, threonine at the 58th amino acid, cannot be found in RA patients and controls in Taiwan. The allele and phenotype frequencies of CYP1A1 4887A and genotype frequency of CYP1A1 4887C/A were lower in RA patients than in controls, whereas the significant difference was lost after correction. MnSOD C1183T polymorphisms were not associated with the clinical manifestations of RA. However, RA patients with CYP1A1 4889G/G have significantly higher frequency of Sjogren's syndrome, especially in the presence of MnSOD 1183T/T. Patients with CYP1A1 4887C/A also have a trend to develop Sjogren's syndrome in the presence of MnSOD 1183T/T. The linkage disequilibrium between CYP1A1 4889G and CYP1A1 6235C can be found in this study. MnSOD gene polymorphisms are not related to susceptibility to RA in Taiwan, whereas individuals with CYP1A1 4887A tend to avoid the development of RA. Moreover, CYP1A1 4889G/G and 4887C/A may play a role in the development of Sjogren's syndrome, especially in the presence of MnSOD 1183T/T. These findings are preliminary. A further confirmation study is necessary.

Yen JH, Chen CJ, Tsai WC, Lin CH, Ou TT, Hu CJ, Liu HW. 2003. Cytochrome P450 and manganese superoxide dismutase genes polymorphisms in systemic lupus erythematosus. Immunol Lett 90(1):19-24.
Abstract: Objectives: To investigate the role of cytochrome P450 1A1 (CYP1A1) and manganese superoxide dismutase (Mn SOD) genes polymorphisms in the pathogenesis of systemic lupus erythematosus (SLE) in Taiwan. Methods: CYP1A1 and Mn SOD genes polymorphisms were determined by the polymerase chain reaction/restriction fragment length polymorphism (RFLP) method in 90 patients with SLE and 94 healthy controls. Results: The genotype frequency of CYP1A1 4887C/A was significantly higher in patients with SLE than in controls. The allele and phenotype frequencies of CYP1A1 4887A were also significantly increased in SLE patients. In contrast, there were no significant differences in the genotype, allele and phenotype frequencies of Mn SOD C1183T polymorphisms between patients with SLE and controls. We also found that SLE patients with CYP1A1 4887A had a trend of increasing prevalence of renal involvement, while Mn SOD C1183T polymorphisms were not associated with the renal involvement in SLE. Conclusions: CYP1A1 4887A may be a precipitating factor for the development of SLE. It also tended to be associated with the occurrence of renal involvement in SLE patients. A synergistic effect was found between CYP1A1 4887C/A and Mn SOD 1183T/T on the susceptibility to SLE. (C) 2003 Elsevier B.V. All rights reserved.

Yang JH, Kang CG, Jin JF, Min NK, Hong SI. 2003. Characterization of Cu-doped PPy based dopamine sensor on n-type silicon substrate. Journal of the Korean Physical Society 42:S542-S546.
Abstract: Dopamine. one of the neurotransmittters in the maininalian central nervous system, plays an important role in early diagnosis of diseases, In the case of a human body, a lower level of dopamine, than the usual causes Parkinson's disease and a higher level causes schizophrenia, The aim of this work is to fabricate a copper (Cu)-doped polypyrrole (PPy) hased dopamine sensitive electrode, A Pt thin-film electrode(1000 Angstrom) is deposited on n-type silicon 1 similar to 10 Ohm(.)cm, orientation: (100)) by embedding an 200 Angstrom Ti underlayer, which increases adhesive strength between the Pt thin-film and silicon dioxide. In general, tyrosinase (EC 1.14, 15.1) is oxidized dopanmine to dopaquinone. Polypyrrole (PPy) films are electropolymerized with Cu on the Pt thin film for increasing conductivity and supporting reagent for oxidation of dopamine. ln addition, tyrosinase. which is included copper in active site, is doped into the Cu-doped PPy film. The sensitivities of a normal PPy-coated dopamine sensitive electrode and the Cu-doped PPY dopamine sensitive electrode are compared by means of chronoamperometry with various dopamine concentrations, Under the optimized condition. the limiting current obtained from the Cu-doped PPY dopamine sensitive electrode is proportional to the dopamine concentrations with the slope of 0.09 muA decade(-1) in the range of 10 mmol/L to 1 mmol/L dopamine concentrations. which is a higher sensitivity value than that of the normal PPy-coated dopamine sensitive electrode, In addition, the electrode surface is analyzed by scanning electron microscopy (SEM) and energy dispersive X-ray spectroscopy (EM).

Yamin G, Glaser CB, Uversky VN, Fink AL. 2003. Certain metals trigger fibrillation of methionine-oxidized alpha-synuclein. J Biol Chem 278(30):27630-27635.
Abstract: The aggregation and fibrillation of alpha-synuclein has been implicated as a key step in the etiology of Parkinson's disease and several other neurodegenerative disorders. In addition, oxidative stress and certain environmental factors, including metals, are believed to play an important role in Parkinson's disease. Previously, we have shown that methionine-oxidized human alpha-synuclein does not fibrillate and also inhibits fibrillation of unmodified alpha-synuclein (Uversky, V. N., Yamin, G., Souillac, P. O., Goers, J., Glaser, C. B., and Fink, A. L. (2002) FEBS Lett. 517, 239-244). Using dynamic light scattering, we show that the inhibition results from stabilization of the monomeric form of Met-oxidized alpha-synuclein. We have now examined the effect of several metals on the structural properties of methionine-oxidized human alpha-synuclein

and its propensity to fibrillate. The presence of metals induced partial folding of both oxidized and non-oxidized alpha-synucleins, which are intrinsically unstructured under conditions of neutral pH. Although the fibrillation of alpha-synuclein was completely inhibited by methionine oxidation, the presence of certain metals (Ti3+, Zn2+, Al3+, and Pb2+) overcame this inhibition. These findings indicate that a combination of oxidative stress and environmental metal pollution could play an important role in triggering the fibrillation of alpha-synuclein and thus possibly Parkinson's disease.

Xie JX, Jiang H, Chen WF, Qian ZM. 2003. Dopamine release rather than content in the caudate putamen is associated with behavioral changes in the iron rat model of Parkinson's disease. Exp Neurol 182(2):483-489.
Abstract: The effects of intranigral iron injection on dopamine (DA) release and content in the caudate putamen (CPu) and their relationship to DA-related behavioral response were investigated in rats. Different concentrations of FeCl3 (10, 20, and 40 mug) and saline were injected separately into the left substantia nigra. In some experiments, rats were pretreated with desferrioxamine or saline before iron injection. After 3 weeks, changes in behavioral response, DA release, and DA content in the CPu were determined. In all iron injection groups (10, 20, and 40 mug), DA content in the lesioned side of the brain was significantly decreased, showing a significant linear correlation (R-2 = 0.981, P < 0.01), and DA turnover ratio significantly increased (both P < 0.01, 0.01 and 0.001 vs unlesioned sides, respectively). However, injection dosages of 10 or 20 mug of iron did not lead to significant changes in DA release in the CPu or in behavioral response. At the 40-mug dosage, it was found that DA release in the lesioned side and rearing activity both were significantly reduced (all P < 0.01 vs unlesioned side or control) and apomorphine-induced rotation was observed. Pretreatment with desferrioxamine significantly inhibited the effect of iron on DA release and content. These results demonstrate that iron injection can damage dopaminergic neurons and suggest that DA release, rather than DA content, in the CPu is associated with DA-related behavioral changes in this PD model. (C) 2003 Elsevier Science (USA). All rights reserved.

Wissler JH. 2003. Redox- and Metalloregulated Rna Bioaptamer Targets of Proteins Associated With Parkinson's and Other Neurodegenerative Diseases - Factors of Relevance in the Life Cycle of CellsVolume 991. p 333-338. Parkinson's Disease: the Life Cycle of the Dopamine Neuron: Annals of the New York Academy of Sciences.

Wang RG, Zhu XZ. 2003. Subtoxic concentration of manganese synergistically potentiates 1-methyl-4-phenylpyridinium-induced neurotoxicity in PC12 cells. Brain Res 961(1):131-138.
Abstract: Endogenous or exogenous substances that are toxic to dopaminergic cells have been proposed as possible cause of idiopathic Parkinson's disease (PD). 1-Methyl-4-phenylpyridinium (MPP+) and manganese are dopaminergic neurotoxins causing a parkinsonism-like syndrome. Here, we studied the possible synergistic reaction between these two neurotoxins using rat PC12 pheochromocytoma cells. MPP+ induced a delayed neurotoxicity in PC12 cells. Although low concentration of manganese did not cause cell damage, it markedly enhanced MPP+-induced neurotoxicity with characteristics of apoptosis, such as DNA laddering and activation of caspase-3. To understand the mechanism of enhancement of subtoxic concentration of manganese on MPP+-induced neurotoxicity, we investigated the reactive oxygen species (ROS) generation using a molecular probe, 2',7'-dichlorofluorescein diacetate. Although subtoxic concentration of manganese alone did not induce ROS increase, it significantly enhanced the ROS generation induced by MPP+. We also determined the intracellular MPP, content. A time- and concentration-dependent increase of MPP+ levels was found in PC12 cells treated with MPP+. The accumulation of MPP+ by PC12 cells was not affected by manganese. Taken together, these studies suggest that co-treatment with MPP+ and manganese may induce synergistic neurotoxicity

in PC12 cells and that subtoxic concentration of manganese may potentiate the effect of MPP+ by an ROS-dependent pathway. (C) 2002 Elsevier Science B.V. All rights reserved.

Wagner KR, Sharp FR, Ardizzone TD, Lu AG, Clark JF. 2003. Heme and iron metabolism: Role in cerebral hemorrhage. J Cereb Blood Flow Metab 23 (6):629-652.
Abstract: Heme and iron metabolism are of considerable interest and importance in normal brain function as well as in neurodegeneration and neuropathologically following traumatic injury and hemorrhagic stroke. After a cerebral hemorrhage, large numbers of hemoglobin-containing red blood cells are released into the brain's parenchyma and/or subarachnoid space. After hemolysis and the subsequent release of heme from hemoglobin, several pathways are employed to transport and metabolize this heme and its iron moiety to protect the brain from potential oxidative stress. Required for these processes are various extracellular and intracellular transporters and storage proteins, the heme oxygenase isozymes and metabolic proteins with differing localizations in the various brain-cell types. In the past several years, additional new genes and proteins have been discovered that are involved in the transport and metabolism of heme and iron in brain and other tissues. These discoveries may provide new insights into neurodegenerative diseases like Alzheimer's, Parkinson's, and Friedrich's ataxia that are associated with accumulation of iron in specific brain regions or in specific organelles. The present review will examine the uptake and metabolism of heme and iron in the brain and will relate these processes to blood removal and to the potential mechanisms underlying brain injury following cerebral hemorrhage.

Vargas JD, Herpers B, Mckie AT, Gledhill S, Mcdonnell J, Van Den Heuvel M, Davies KE, Ponting CP. 2003. Stromal cell-derived receptor 2 and cytochrome b561 are functional ferric reductases. Biochimica Et Biophysica Acta-Proteins and Proteomics 1651(1-2):116-123.
Abstract: Iron has a variety of functions in cellular organisms ranging from electron transport and DNA synthesis to adenosine triphosphate (ATP) and neurotransmitter synthesis. Failure to regulate the homeostasis of iron can lead to cognition and demyelination disorders when iron levels are deficient, and to neurodegenerative disorders when iron is in excess. In this study we show that three members of the b561 family of predicted ferric reductases, namely mouse cytochrome b561 and mouse and fly stromal cell-derived receptor 2 (SDR2), have ferric reductase activity. Given that a fourth member, duodenal cytochrome b (Dcytb), has previously been shown to be a ferric reductase, it is likely that all remaining members of this family also exhibit this activity. Furthermore, we show that the rat sdr2 message is predominantly expressed in the liver and kidney, with low expression in the duodenum. In hypotransferrinaemic (hpx) mice, sdr2 expression in the liver and kidney is reduced, suggesting that it may be regulated by iron. Moreover, we demonstrate the presence of mouse sdr2 in the choroid plexus and in the ependymal cells lining the four ventricles, through in situ hybridization analysis. (C) 2003 Elsevier B.V. All rights reserved.

Turubull S, Tabner BJ, Brown DR, Allsop D. 2003. Generation of hydrogen peroxide from mutant forms of the prion protein fragment PrP121-231. Biochemistry (Mosc) 42(25):7675-7681.
Abstract: By means of electron spin resonance spectroscopy, in conjunction with the spin trapping technique, we have shown previously that Abeta and alpha-synuclein (aggregating proteins that accumulate in the brain in Alzheimer's disease, Parkinson's disease, and related disorders) both induce the formation of hydroxyl radicals following incubation in solution, upon addition of Fe(II). These hydroxyl radicals are apparently formed from hydrogen peroxide, via Fenton's reaction. An N-terminally truncated fragment of the mouse prion protein (termed PrP121-231) is toxic to cerebellar cells in culture, and certain human mutations, responsible for inherited prion disease, enhance this toxicity.

Here we report that PrP121-231 containing three such mutations (E200K, D178N, and F198S) also generated hydroxyl radicals, upon addition of Fe (11). The formation of these radicals was blocked by catalase, or by metal chelators, each of which also reduced the toxicity of the PrP121-231 fragments to cultured normal mouse cerebellar cells. Wild-type PrP121-231, full-length cellular PrP, and its homologue doppel did not generate any detectable hydroxyl radicals. We conclude that the additional cytotoxic effects of the mutant forms of PrP121-231 could be due to their ability to generate hydrogen peroxide, by a metal-dependent mechanism. Thus, one effect of these (and possibly other) prion mutations could be production of a particularly toxic form of the prion protein, with an enhanced capacity to induce oxidative damage, neurodegeneration, and cell loss.

Tsang F, Soong TW. 2003. Interactions between environmental and genetic factors in the pathophysiology of Parkinson's disease. Iubmb Life 55(6): 323-327.
Abstract: Parkinson's disease (PD) is a progressive neurodegenerative disease with no known cure and affects approximately 1% of the elderly population. The major question in PD relates to the selective loss of dopaminergic neurons in patients. The underlying mechanism of genetic dysfunction and environmental toxins in contributing to the pathogenesis of PD may be oxidative stress. The interactions of genetic and environmental factors in PD may provide some answers to the longstanding question. In particular, the possibility that iron may provide selectivity to genetic susceptibility or dopamine reactivity in dopaminergic neuronal death is enhanced by the neuroprotection demonstrated in transgenic mice overexpressing ferritin or the use of iron chelators in MPTP-induced PD mouse. It will be important to dissect and understand the contributions of genes, environment and intrinsic cellular states in the generation and progression of the pathophysiology of PD.

Thompson K, Menzies S, Muckenthaler M, Torti FM, Wood T, Torti SV, Hentze MW, Beard J, Connor J. 2003. Mouse brains deficient in H-ferritin have normal iron concentration but a protein profile of iron deficiency and increased evidence of oxidative stress. J Neurosci Res 71(1):46-63.
Abstract: Several neurodegenerative disorders such as Parkinson's Disease (PD) and Alzheimer's Disease (AD) are associated with elevated brain iron accumulation relative to the amount of ferritin, the intracellular iron storage protein. The accumulation of more iron than can be adequately stored in ferritin creates an environment of oxidative stress. We developed a heavy chain (H) ferritin null mutant in an attempt to mimic the iron milieu of the brain in AD and PD. Animals homozygous for the mutation die in utero but the heterozygotes (+/-) are viable. We examined heterozygous and wild-type (wt) mice between 6 and 8 months of age. Macroscopically, the brains of +/- mice were well formed and did not differ from control brains. There was no evidence of histopathology in the brains of the heterozygous mice. Iron levels in the brain of the +/- and wild-type (+/+) mice were similar, but +/- mice had less than half the levels of H-ferritin. The other iron management proteins transferrin, transferrin receptor, light chain ferrittin, Divalent Metal Transporter 1, ceruloplasmin, were increased in the +/- mice compared to +/+ mice. The relative amounts of these proteins in relation to the iron concentration are similar to that found in AD and PD. Thus, we hypothesized that the brains of the heterozygote mice should have an increase in indices of oxidative stress. In support of this hypothesis, there was a decrease in total superoxide dismutase (SOD) activity in the heterozygotes coupled with an increase in oxidatively modified proteins. In addition, apoptotic markers Bax and caspase-3 were detected in neurons of the +/- mice but not in the wt. Thus, we have developed a mouse model that mimics the protein profile for iron management seen in AD and PD that also shows evidence of oxidative stress. These results suggest that this mouse may be a model to determine the role of iron mismanagement in neurodegenerative disorders and for testing antioxidant therapeutic strategies. (C) 2002 Wiley-Liss, Inc.

Tan EK, Tan C, Fook-Chong SMC, Lum SY, Chai A, Chung H, Shen H, Zhao Y, Teoh ML, Yih Y, Pavanni R, Chandran VR, Wong MC. 2003. Dose-dependent protective effect of coffee, tea, and smoking in Parkinson's disease: a study in ethnic Chinese. J Neurol Sci 216(1):163-167.
Abstract: Introduction: Few studies have examined the relationship of coffee and tea in Parkinson's disease (PD). The potential protective effect of coffee intake and risk of PD has not been studied in a Chinese population. There is a high prevalence of caffeine takers among Chinese in our population. Objective: We undertook a case control study to examine the relationship between coffee and tea drinking, cigarette smoking, and other enviromental factors and risk of PD among ethnic Chinese in our population. Methods and Results: 300 PD and 500 population controls were initially screened. Two hundred case control pairs matched for age, gender, and race were finally included in the analysis. Univariate analysis revealed significant association of PD with coffee drinking ($p<0.0005$), tea drinking ($p=0.019$), alcohol drinking ($p=0.001$), cigarette smoking ($p<0.0005$), and exposure to heavy metals ($p=0.006$). Conditional logistic regression analysis demonstrated that amount of coffee drunk (OR 0.787, 95%CI 0.664-0.932, $p=0.006$), amount of tea drunk (OR 0.724, 95%CI 0.559-0.937, $p=0.014$), number of cigarettes smoked (OR 0.384,95%CI 0.204-0.722,$p = 0.003$), history of heavy metal and toxin exposure (OR 11.837,95%CI 1.075-130.366, $p=0.044$), and heart disease (OR 5.518, 95%CI 1.377-22.116, $p=0.016$) to be significant factors associated with PD. One unit of coffee and tea (3 cups/day for 10 years) would lead to a 22% and 28% risk reduction of PD. One unit of cigarette smoke (3 packs/day for 10 years) reduced the risk of PD by 62%. Conclusions: We demonstrated a dose-dependent protective effect of PD in coffee and tea drinkers and smokers in an ethnic Chinese population. A history of exposure to heavy metals increased the risk of PD, supporting the multifactorial etiologies of the disease. (C) 2003 Elsevier B.V. All rights reserved.

Tan DX, Manchester LC, Sainz R, Mayo JC, Alvares FL, Reiter RJ. 2003. Antioxidant strategies in protection against neurodegenerative disorders. Expert Opinion on Therapeutic Patents 13(10):1513-1543.
Abstract: The most common neurodegenerative diseases include Alzheimer's disease, Parkinson's disease and stroke; they are devastating clinical problems which lack effective treatments. Although the aetiology of these diseases is not fully understood, oxidative stress is believed to be a contributing causative factor. In addition to conventional therapies, antioxidant strategies in protection against neurodegenerative conditions have been increasingly addressed, as evidenced by an increasing number of animal studies, clinical reports and patents regarding these processes in recent years. The effectiveness of antioxidants in protecting against neurodegenerative disorders lies mainly in their ability to cross the blood-brain barrier, their potential in terms of subcellular distribution occurring in membranes, in the cytoplasm and especially in mitochondria, and their multifunctional capacity as well as their synergistic actions. The naturally occurring antioxidants with different properties collaborate as an array to defend against oxidative stress. Single antioxidant supplementation would not then be expected to have a remarkable influence on neurodegenerative diseases, which may involve free radicals. Thus, using combinations of antioxidants with different subcellular distributions and different properties for prophylaxis or treatment would probably improve therapeutic outcomes. Based on their multifactoral aetiology, the development of novel antioxiclants with anti-inflammatory and metal-chelating properties and the ability to improve metabolism, for example by increasing ATP production rate or a new formulation of antioxiclants with other agents, which have different functions, will become the new strategies in protecting against neurodegenerative disorders.

Takser L, Mergler D, Hellier G, Sahuquillo J, Huel G. 2003. Manganese, monoamine metabolite levels at birth, and child psychomotor development. Neurotoxicology 24(4-5):667-674.
Abstract: Several studies have demonstrated neurobehavioral impairment

related to manganese (Mn) exposure in the workplace. Exposure to high doses of manganese is associated with irreversible neurodegenerative disorders resembling idiopathic Parkinson disease. Although there is a risk of Mn accumulation in the foetus during pregnancy, little information exists about developmental effects of environmental low-level exposure in human. For this reason, we conducted a prospective epidemiological study in 247 healthy pregnant women and their babies to determine the long-term effect of in utero Mn levels on child's psychomotor development. Concurrently, we examined the relationship between Mn tissue levels at delivery and foetal plasma monoamine metabolites. Of the newborns, 195 were examined at 9 months, 126 at 3 years and 100 at 6 years. At 9 months, the Brunet-Lezine scales were administered. The McCarthy scales of children's abilities were used at 3 and 6 years. After adjustment for potential confounding co-factors (child's gender, mother's educational level), negative relationships were observed between cord blood Mn levels and several psychomotor sub-scales at age of 3 years: attention (partial r = -0.33, P < 0.001), non-verbal memory (partial r = -0.28, P < 0.01), and hand skills (partial r = -0.22, P < 0.05). No significant relationships were observed between Mn measures at birth and the general psychomotor indices, Brunet-Lezine developmental quotient (DQ) at 9 months or McCarthy general cognitive index (GCI) at 3 and 6 years. Maternal blood Mn levels were negatively associated with foetal plasma HVA and 5-HIAA concentrations (adjusted for labour duration, child's gender, and smoking during pregnancy), but the adjustment for monoamine levels at birth did not change the association between the Mn levels and the psychomotor scores. These results suggest that environmental Mn exposure in utero could affect early psychomotor development. (C) 2003 Elsevier Science Inc. All rights reserved.

Takeda A. 2003. Manganese action in brain function. Brain Research Reviews 41 (1):79-87.
Abstract: Manganese, an essential trace metal, is supplied to the brain via both the blood-brain and the blood-cerebrospinal fluid barriers. There are some mechanisms in this process and transferrin may be involved in manganese transport into the brain. A large portion of manganese is bound to manganese metalloproteins, especially glutamine synthetase in astrocytes. A portion of manganese probably exists in the synaptic vesicles in glutamatergic neurons and the manganese is dynamically coupled to the electrophysiological activity of the neurons. Manganese released into the synaptic cleft may influence synaptic neurotransmission. Dietary manganese deficiency, which may enhance susceptibility to epileptic functions, appears to affect manganese homeostasis in the brain, probably followed by alteration of neural activity. On the other hand, manganese also acts as a toxicant to the brain because this metal has prooxidant activity. Abnormal concentrations of manganese in the brain, especially in the basal ganglia, are associated with neurological disorders similar to Parkinson's disease. Understanding the movement and action of manganese in synapses may be important to clarify the function and toxicity of manganese in the brain. (C) 2002 Elsevier Science B.V. All rights reserved.

Szczerbowska-Boruchowska M, Lankosz M, Ostachowicz J, Adamek D, Krygowska-Wajs A, Tomik B, Szczudlik A, Simionovici A, Bohic S. 2003. Application of synchrotron radiation for elemental microanalysis of human central nervous system tissue. Journal De Physique Iv 104:325-328.
Abstract: The pathogenesis of two neurodegenerative diseases i.e. Parkinson's Disease (PD) and amyotrophic lateral sclerosis (ALS) are still not known. It is supposed that disturbance of metal ions homeostasis may promote degeneration and atrophy of neurones. As a preliminary study, the quantitative and topographic elemental analysis of selected parts of human brain and spinal cord was performed using synchrotron microbeam-X ray fluorescence (mu-SXRF) technique. The samples were taken during the autopsy from patients with PD, ALS and from patients died due to non-neurological conditions events. X-ray fluorescence imaging showed that increased concentration of selected elements are observed in neurons

perikarial parts in compare with surrounding area. Moreover, comparable analysis showed significant differences in accumulation of selected elements between the pathological and control cases. The investigations indicate that micro-beam of synchrotron radiation can be satisfactory applied for analysis of central nervous system tissue providing useful information about distribution and contents of elements at the single cell level.

Surgucheva IG, Sivak JM, Fini ME, Palazzo RE, Surguchov AP. 2003. Effect of gamma-synuclein overexpression on matrix metalloproteinases in retinoblastoma Y79 cells. Arch Biochem Biophys 410(1):167-176.
Abstract: gamma-Synuclein is a small cytoplasmic protein implicated in neurodegenerative diseases and cancer. However, the mechanism of its involvement in diseases is not clear. We studied the role of gamma-synuclein in the regulation of matrix metalloproteinases in retinoblastoma cell culture. Matrix metalloproteinases play important roles in the remodeling of extracellular matrix implicated in tumor progression and in the neurodegenerative diseases. Western blot and zymography data demonstrated a moderate elevation of matrix metalloproteinases-2 and significant upregulation of matrix metalloproteinases-9 in stable cell lines overexpressing gamma-synuclein. No effect of gamma-synuclein overexpression on matrix metalloproteinases-1 level or activity was found. Chloramphenicol-acetyltransferase assay demonstrated that overexpression of gamma-synuclein increases the efficiency of the matrix metalloproteinases-9 promoter. This increment of promoter activity may be mediated by the AP-1 binding site(s), since point mutations in one of these sites (Pr18 or Pr19) and elimination of the distal AP-1 site (Pr14) reduced the increment of promoter activity. (C) 2002 Elsevier Science (USA). All rights reserved.

Soh Y, Shin MH, Lee JS, Jang JH, Kim OH, Kang H, Surh YJ. 2003. Oxidative DNA damage and glioma cell death induced by tetrahydropapaveroline. Mutation Research-Reviews in Mutation Research 544(2-3):129-142.
Abstract: A series of naturally occurring isoquinoline alkaloids, besides their distribution in the environment and presence in certain food stuffs, have been detected in human tissues including particular regions of brain. An example is salsolinol (1-methyl-6,7-dihydroxy-1,2,3,4-tetrahydroisoquinoline) that not only induces neuronal cell death, but also causes DNA damage and genotoxicity. Tetrahydropapaveroline [THP; 6,7-dihydroxy-1-(3',4'-dihydroxybenzyl)-1,2,3,4-tetrahydroisoquinoline], a dopamine-derived tetrahydroisoquinoline alkaloid, has been reported to inhibit mitochondrial respiration and is considered to contribute to neurodegeneration implicated in Parkinson's disease. Since THP bears two catechol moieties, the compound may readily undergo redox cycling to produce reactive oxygen species (ROS) as well as toxic quinoids. In the present study, we have examined the capability of THP to cause oxidative DNA damage and cell death. Incubation of THP with phiX174 supercoiled DNA or calf thymus DNA in the presence of cupric ion caused substantial DNA damage as determined by strand scission or formation of 8-oxo-7,8-dihydro-2'-deoxyguanosine (8-oxodGuo), respectively. THP plus copper-induced DNA damage was ameliorated by some ROS scavengers/antioxidants and catalase. Treatment of C6 glioma cells with THP led to a concentration-dependent reduction in cell viability, which was prevented by the antioxidant N-acetyl-L-cysteine. When these cells were treated with 10 muM THP, c-Jun N-terminal kinase (INK) and p38 mitogen-activated protein kinase (MAPK) were rapidly activated via phosphorylation, whereas activation of extracellular signal-regulated protein kinase (ERK) was inhibited. Furthermore, pretreatment with inhibitors of INK and p38 MAPK rescued the glioma cells from THP-induced cytotoxicity, suggestive of the involvement of these kinases in THP-induced C6 glioma cell damage. (C) 2003 Elsevier B.V. All rights reserved.

Sharma SK, Ebadi M. 2003. Metallothionein attenuates 3-morpholinosydnonimine (SIN-1)-induced oxidative stress in dopaminergic neurons. Antioxidants & Redox Signaling 5(3):251-264.

Abstract: Parkinson's disease is characterized by a progressive loss of dopaminergic neurons in the substantia nigra. zona compacta, and in other subcortical nuclei associated with a widespread occurrence of Lewy bodies. The causes of cell death in Parkinson's disease are still poorly understood, but a defect in mitochondrial oxidative phosphorylation and enhanced oxidative stress have been proposed. We have examined 3-morpholinosydnonimine (SIN-1)-induced apoptosis in control and metallothionein-overexpressing dopaminergic neurons, with a primary objective to determine the neuroprotective potential of metallothionein against peroxynitrite-induced neurodegeneration in Parkinson's disease. SIN-I induced lipid peroxidation and triggered plasma membrane blebbing. In addition, it caused DNA fragmentation, alpha-synuclein induction, and intramitochondrial accumulation of metal ions (copper, iron, zinc, and calcium), and enhanced the synthesis of 8-hydroxy-2-deoxyguanosine. Furthermore, it down-regulated the expression of Bcl-2 and poly(ADP-ribose) polymerase, but up-regulated the expression of caspase-3 and Bax in dopaminergic (SK-N-SH) neurons. SIN-1 induced apoptosis in aging mitochondrial genome knockout cells, alpha-synuclein-transfected cells, metallothionein double-knockout cells, and caspase-3-overexpressed dopaminergic neurons. SIN-1-induced changes were attenuated with selegiline or in metallothionein-transgenic striatal fetal stem cells. SIN-1-induced oxidation of dopamine to dihydroxyphenylacetaldehyde was attenuated in metallothionein-transgenic fetal stem cells and in cells transfected with a mitochondrial genome, and enhanced in aging mitochondrial genome knockout cells, in metallothionein double-knockout cells and caspase-3 gene-overexpressing dopaminergic neurons. Selegiline, melatonin, ubiquinone, and metallothionein suppressed SIN-1-induced down-regulation of a mitochondrial genome and up-regulation of caspase-3 as determined by reverse transcription-polymerase chain reaction. The synthesis of mitochondrial 8-hydroxy-2-deoxyguanosine and apoptosis-inducing factors were increased following exposure to 1-methyl-4-phenylpyridinium ion or rotenone. Pretreatment with selegiline or metallothionein suppressed 1-methyl-4-phenylpyridinium ion-, 6-hydroxydopamine-, and rotenone-induced increases in mitochondrial 8-hydroxy-2-deoxyguanosine accumulation. Transfection of aging mitochondrial genome knockout neurons with mitochondrial genome encoding complex-1 or melanin attenuated the SIN-1-induced increase in lipid peroxidation. SIN-1 induced the expression of alpha-synuclein, caspase-3, and 8-hydroxy-2-deoxyguanosine, and augmented protein nitration. These effects were attenuated by metallothionein gene overexpression. These studies provide evidence that nitric oxide synthase activation and peroxynitrite ion overproduction may be involved in the etiopathogenesis of Parkinson's disease, and that metallothionein gene induction may provide neuroprotection.

Savoiardo M. 2003. Differential diagnosis of Parkinson's disease and atypical parkinsonian disorders by magnetic resonance imaging. Neurological Sciences 24:S35-S37.
Abstract: Magnetic resonance imaging (MRI) is not usually considered helpful for establishing the diagnosis of Parkinson's disease, but it is much more important to support the clinical diagnosis in atypical parkinsonian disorders. In multiple system atrophy with predominant parkinsonian features (MSA-P), MRI demonstrates putaminal abnormalities due to loss of neurons and gliosis and accumulation of iron in the posterior lateral part of the nucleus. When cerebellar features are present (MSA-C), pontine and cerebellar atrophy is seen with signal abnormalities that correspond to the distribution of the degenerative changes. In progressive supranuclear palsy, the main abnormality is atrophy of the midbrain. Mild-to-moderate cerebral atrophy may be present, but more-marked asymmetrical atrophy in the posterior frontal and parietal regions contralateral to the side of the clinical manifestations is characteristic of corticobasal degeneration.

Sadek AH, Rauch R, Schulz PE. 2003. Parkinsonism due to Manganism in a Welder. International Journal of Toxicology 22(5):393-401.
Abstract: A 33-year-old right-handed male presented complaining of a 2-

year history of progressive cognitive slowing, rigidity, tremors, slowing of movements, and gait instability leading to falls. On examination, he had a Mini-Mental Status Examination (MMSE) score of 29, slowed saccadic eye pursuit, hypomimia, cogwheel rigidity, a 3- to 4-Hz tremor, and a "cock-walk" gait. His symptoms and signs were similar to idiopathic Parkinson's disease; however, he was young, inattention and forgetfulness occurred early in the course of the disorder, levodopa was unhelpful, and his gait was atypical. His work up for secondary causes of parkinsonism was negative, except for increased signal intensity on T1-weighted magnetic resonance image (MRI) in the bilateral basal ganglia. Typical etiologies for that finding were ruled-out, which led to further inquiries into the patient's lifestyle. He was a welder, and discussion with his employer revealed that he used a steel-manganese alloy, he often worked in a confined ship's hold, and he did not use a respiratory mask. Because manganese toxicity can produce increased T1-weighted signal intensities in the basal ganglia, the authors tested his serum and urine manganese, and both were elevated. This patient emphasizes the importance of a careful occupational history in persons presenting with atypical manifestations of a neurodegenerative disorder. It also lends support to the hypothesis that welding can produce enough exposure to manganese to produce neurologic impairment.

Ruprecht-Dorfler P, Berg D, Tucha O, Benz P, Meier-Meitinger M, Alders GL, Lange KW, Becker G. 2003. Echogenicity of the substantia nigra in relatives of patients with sporadic Parkinson's disease. Neuroimage 18(2):416-422.
Abstract: Increased echogenicity of the substantia nigra (SN) on ultrasound is a typical sonograpbic finding in Parkinson's disease (PD). Sonographic signal intensity of the SN is related to tissue iron content with higher iron level being associated with increased echogenicity. Recent findings indicate that hyperechogenicity of the SN represents an important susceptibility factor for nigrostriatal degeneration. In this study we determined the prevalence of a characteristic ultrasound sign of Parkinson's disease in first-degree relatives of PD patients. Fourteen patients with sporadic PD and 58 of their relatives underwent neurological, neuropsychological, and ultrasound examination. In addition, four pairs of relatives (one member of each pair exhibiting increased echogenicity of the SN and the other with regular SN echogenicity) underwent 18 F-Dopa PET examination. On transcranial sonography, 26 of the 58 relatives exhibited SN hyperechogenicity. Twenty-four relatives showed minor signs of motor slowing. Relatives with SN hyperechogenicity more often showed signs of hypokinesia (16 v 8 relatives; U test, P = 0.01) and impaired executive functions (Tower of London task, problems solved with the minimum number of moves; U test, P = 0.012) than relatives without this echo pattern. In addition, F-18-Dopa uptake (influx constants) at the putamen was reduced in subjects with SN hyperechogenicity compared to their relatives without this ultrasound sign (Wilcoxon, P = 0.03). In conclusion, approximately 45% of relatives of PD patients exhibited an increased echogenicity of the SN. This sign is associated with clinical findings and objective measurements, indicating some degree of impaired nigrostriatal function. (C) 2003 Elsevier Science (USA). All rights reserved.

Ruggera PS, Witters DM, Von Maltzahn G, Bassen HI. 2003. In vitro assessment of tissue heating near metallic medical implants by exposure to pulsed radio frequency diathermy. Phys Med Biol 48(17):2919-2928.
Abstract: A patient with bilateral implanted neurostimulators suffered significant brain tissue damage, and subsequently died, following diathermy treatment to hasten recovery from teeth extraction. Subsequent MRI examinations showed acute deterioration of the tissue near the deep brain stimulator (DBS) lead's electrodes which was attributed to excessive tissue heating induced by the diathermy treatment. Though not published in the open literature, a second incident was reported for a patient with implanted neurostimulators for the treatment of Parkinson's disease. During a diathermy treatment for severe kyphosis, the patient had a sudden change in mental status and neurological deficits. The diathermy was implicated in causing damage to the patient's brain tissue. To

investigate if diathermy induced excessive heating was possible with other types of implantable lead systems, or metallic implants in general, we conducted a series of in vitro laboratory tests. We obtained a diathermy unit and also assembled a controllable laboratory exposure system. Specific absorption rate (SAR) measurements were performed using fibre optic thermometry in proximity to the implants to determine the rate of temperature rise using typical diathermy treatment power levels. Comparisons were made of the SAR measurements for a spinal cord stimulator (SCS) lead, a pacemaker lead and three types of bone prosthesis (screws, rods and a plate). Findings indicate that temperature changes of 2.54 and 4.88 degreesC s(-1) with corresponding SAR values of 9129 and 17 563 W kg(-1) near the SCS and pacemaker electrodes are significantly higher than those found in the proximity of the other metallic implants which ranged from 0.04 to 0.69 degreesC s(-1) (129 to 2471 W kg (-1)). Since the DBS leads that were implanted in the reported human incidents have one-half the electrode surface area of the tested SCS lead, these results imply that tissue heating at rates at least equal to or up to twice as much as those reported here for the SCS lead could occur for the DBS leads.

Roth JA, Garrick MD. 2003. Iron interactions and other biological reactions mediating the physiological and toxic actions of manganese. Biochem Pharmacol 66(1):1-13.
Abstract: Chronic exposure to the divalent heavy metals, such as iron, lead, manganese (Mn), and chromium, has been linked to the development of severe, often irreversible neurological disorders and increased vulnerability to developing Parkinson's disease. Although the mechanisms by which these metals elicit or facilitate neuronal cell death are not well defined, neurotoxicity is limited by the extent to which they are transported across the blood-brain barrier and their subsequent uptake within targeted neurons. Once inside the neuron, these heavy metals provoke a series of biochemical and molecular events leading to cell death induced by either apoptosis and/or necrosis. The toxicological properties of Mn have been studied extensively in recent years because of the potential health risk created by increased atmospheric levels owing to the impending use of the gas additive methylcyclopentadienyl manganese tricarbonyl. Individuals exposed to high environmental levels of Mn, which include miners, welders, and those living near ferroalloy processing plants, display a syndrome known as manganism, best characterized by debilitating symptoms resembling those of Parkinson's disease. Mn disposition in vivo is influenced by dietary iron intake and stores within the body since the two metals compete for the same binding protein in serum (transferrin) and subsequent transport systems (divalent metal transporter, DMTI). There appear to be two distinct carrier-mediated transport systems for Mn and ferrous ion: a transferrin-dependent and a transferrin-independent pathway, both of which utilize DMT1 as the transport protein. Accordingly, this commentary focuses on the biochemical and molecular processes responsible for the cytotoxic actions of Mn and the role that cellular transport plays in mediating the physiological as well as the toxicological actions of this metal. (C) 2003 Elsevier Science Inc. All rights reserved.

Ren MQ, Ong WY, Wang XS, Watt F. 2003. A nuclear microscopic and histochemical study of iron concentrations and distribution in the midbrain of two age groups of monkeys unilaterally injected with MPTP. Exp Neurol 184(2):947-954.
Abstract: The present study was carried out to elucidate the concentration and distribution of iron in the substantia nigra of two age groups of monkeys after experimental hemi-Parkinsonism induced by unilateral internal carotid injections of MPTP. Iron levels and distribution were detected using the nuclear microscope, which is able to provide structural and quantitative elemental analysis of biological tissue down to the parts per million (ppm) level of analytical sensitivity. Five weeks after unilateral lesioning with MPTP, we observed a 30-65% loss of neurons in the injected substantia nigra of each monkey, compared with the contralateral control 'non-lesioned' side. In monkeys less than 7 years of age, the iron was

distributed fairly uniformly and showed little evidence of focal deposits. In monkeys greater than 7 years of age, we observed many dense focal deposits of iron in the substantia nigra. A comparison between iron distributions in nuclear microscopic scans and cell distributions in the same sections stained by the Nissl technique showed that areas containing high iron concentrations were present not where large-diameter neurons with abundant Nissl substance (presumed dopaminergic neurons) were located but in a region ventral to these cell bodies, i.e., in the substantia nigra pars reticulata. These distributions were present on the control side as well as the MPTP-injected side. Since a previous study has shown that unilateral MPTP injection results in lesions of the substantia nigra of the same side but negligible injury to the opposite side, this implies that the iron deposits existed in the older monkeys before MPTP injections (i.e. they occurred normally). The accumulation of iron in the substantia nigra with age suggests the possibility of localised damage to neurons through the catalysis of free radicals. (C) 2003 Elsevier Inc. All rights reserved.

Reich SG, Malecki E, Moliterno AR, Corse AM, Lee LA, Vogelsang GB. 2003. Manganese-induced parkinsonism from total parenteral nutrition: Report of a case and review of the literature. Ann Neurol 54:S73.

Powers KM, Smith-Weller T, Franklin GM, Longstreth WT, Swanson PD, Checkoway H. 2003. Parkinson's disease risks associated with dietary iron, manganese, and other nutrient intakes. Neurology 60(11):1761-1766.
Abstract: Background: Dietary influences on oxidative stress have been thought to play important role in the etiology of PD. Objective: To examine associations of PD with dietary nutrients, including minerals, vitamins, and fats. Methods: A population-based case-control study was conducted among newly diagnosed case (n = 250) and control subjects (n = 388) identified between 1992 and 2002 from enrollees of the Group Health Cooperative health maintenance organization in western Washington state. Controls were frequency matched to cases on sex and age. In-person interviews elicited data on food frequency habits during most of adult life. Nutrient intakes were calculated and analyzed by adjusting each person's nutrient intake by their total energy intake (the nutrient density technique). Results: Subjects with an iron intake in the highest quartile compared with those in the lowest quartile had an increased risk of PD (odds ratio = 1.7, 95% CI: 1.0, 2.7, trend p = 0.016). There was an apparent joint effect of iron and manganese; dietary intake above median levels of both together conferred a nearly doubled risk compared with lower intakes of each nutrient (odds ratio = 1.9, 95% CI: 1.2, 2.9). No strong associations were found for either antioxidants or fats. Conclusion: A high intake of iron, especially in combination with high manganese intake, may be related to risk for PD.

Pardini C, Vaglini F, Galimberti S, Corsini GU. 2003. Dose-dependent induction of apoptosis by R-apornorphine in CHO-K1 cell line in culture. Neuropharmacology 45(2):182-189.
Abstract: A variety of mechanisms have been proposed as explanations for the distinctive neuropathology of Parkinson's disease, such as increased iron levels, increased oxidant stress or decreased antioxidant defences. The vulnerability of dopamine-containing neurons towards cell death has attracted much attention to the dopamine molecule itself as one of the probable neurotoxic factors leading to neurodegeneration. The similarity between apomorphine and dopamine with regards to their chemical, pharmacological and toxicological properties provided a basis for investigating the nature of the toxicity of the former agent. In this study the CHO-K1 cell line was exposed to different concentrations of apomorphine, and markers of cell death and apoptosis were studied. Apomorphine reduced cell proliferation in a dose-dependent fashion after 72 h incubation. Furthermore, apomorphine induced dose-dependent cell death at concentrations of 10-50 muM. The CHO-K1 line showed specific markers of apoptosis such as the typical DNA laddering phenomenon on agarose gel, morphological changes of apoptotic nuclei as described by in situ end labelling, and annexin binding. These data strongly suggest that

apomorphine, like dopamine, elicits its cytotoxic effect with an apoptotic mechanism. (C) 2003 Elsevier Science Ltd. All rights reserved.

Pan TH, Jankovic J, Le WD. 2003. Potential therapeutic properties of green tea polyphenols in Parkinson's disease. Drugs & Aging 20(10):711-721.
Abstract: Tea is one of the most frequently consumed beverages in the world. It is rich in polyphenols, a group of compounds that exhibit numerous biochemical activities. Green tea is not fermented and contains more catechins than black tea or oolong tea. Although clinical evidence is still limited, the circumstantial data from several recent studies suggest that green tea polyphenols may promote health and reduce disease occurrence, and possibly protect against Parkinson's disease and other neurodegenerative diseases. Green tea polyphenols have demonstrated neuroprotectant activity in cell cultures and animal models, such as the prevention of neurotoxin-induced cell injury. The biological properties of green tea polyphenols reported in the literature include antioxidant actions, free radical scavenging, iron-chelating properties, H-3-dopamine and H-3-methyl-4-phenylpyridine uptake inhibition, catechol-O-methyl-transferase activity reduction, protein kinase C or extracellular signal-regulated kinases signal pathway activation, and cell survival/cell cycle gene modulation. All of these biological effects may benefit patients with Parkinson's disease. Despite numerous studies in recent years, the understanding of the biological activities and health benefits of green tea polyphenols is still very limited. Further in-depth studies are needed to investigate the safety and efficacy of green tea in humans and to determine the different mechanisms of green tea in neuroprotection.

Pals P, Van Everbroeck B, Grubben B, Viaene MK, Dom R, Van Der Linden C, Santens P, Martin JJ, Cras P. 2003. Case-control study of environmental risk factors for Parkinson's disease in Belgium. Eur J Epidemiol 18(12): 1133-1142.
Abstract: The aetiology of Parkinson's disease (PD) is unknown and said to be multifactorial. We report on a retrospective epidemiological case control study, performed in Flanders during a 3- year period, investigating known and potential environmental risk factors for PD by means of questionnaires. We investigated 423 prevalent patients and 205 spouse-controls. We found familial occurrence in 15% of the patients, a mean age of onset of 58 years, and a clear male preponderance (male/ female ratio 1.53). Our results suggest more nulliparity among female PD patients (95% CI: 1.08 - 5.76). We found a discrete clustering of patients in areas with intensive metallurgic activity. Patients were more frequently employed in metallurgy than controls (95% CI: 1.04 - 9.20). Furthermore, patients were clearly more exposed to zinc (95% CI: 1.51 - 90.90) and toluene (95% CI: 1.03 - 58.82). Male patients report more prostatectomy- surgery (95% CI: 1.54 - 17.24).

Palomo T, Beninger RJ, Kostrzewa RM, Archer T. 2003. Brain sites of movement disorder: Genetic and environmental agents in neurodevelopmental perturbations. Neurotoxicity Research 5(1-2):1-26.
Abstract: In assessing and assimilating the neurodevelopmental basis of the so-called movement disorders it is probably useful to establish certain concepts that will modulate both the variation and selection of affliction, mechanisms-processes and diversity of disease states. Both genetic, developmental and degenerative aberrations are to be encompassed within such an approach, as well as all deviations from the necessary components of behaviour that are generally understood to incorporate "normal" functioning. In the present treatise, both conditions of hyperactivity/ hypoactivity, akinesia and bradykinesia together with a constellation of other symptoms and syndromes are considered in conjunction with the neuropharmacological and brain morphological alterations that may or may not accompany them, e.g. following neonatal denervation. As a case in point, the neuroanatomical and neurochemical points of interaction in Attention Deficit and Hyperactivity disorder (ADHD) are examined with reference to both the perinatal metallic and organic environment and genetic backgrounds. The role of apoptosis, as opposed to necrosis, in cell

death during brain development necessitates careful considerations of the current explosion of evidence for brain nerve growth factors, neurotrophins and cytokines, and the processes regulating their appearance, release and fate. Some of these processes may possess putative inherited characteristics, like a-synudein, others may to greater or lesser extents be endogenous or semi-endogenous (in food), like the tetrahydroisoquinolines, others exogenous until inhaled or injested through environmental accident, like heavy metals, e.g. mercury. Another central concept of neurodevelopment is cellular plasticity, thereby underlining the essential involvement of glutamate systems and N-methyl-D-aspartate receptor configurations. Finally, an essential assimilation of brain development in disease must delineate the relative merits of inherited as opposed to environmental risks not only for the commonly regarded movement disorders, like Parkinson's disease, Huntington's disease and epilepsy, but also for afflictions bearing strong elements of psychosocial tragedy, like ADHD, autism and Savantism.

Ogasawara Y, Ohata E, Sakamoto T, Ishii K, Takahashi H, Tanabe S. 2003. A model of aluminum exposure associated with lipid peroxidation in rat brain. Biol Trace Elem Res 96(1-3):191-201.
Abstract: We have developed a rat model to investigate the relationship between aluminum exposure and aluminum accumulation, and with oxidative damage in brain tissues. Intraperitoneal injections of aluminum lactate for 7 wk (the total aluminum dosage per rat was approx 100 mg) significantly increased aluminum levels in the brain. The concentration of lipid peroxidation products (thiobarbituric acid-reactive substances [TBARS]) also increased in the brain following aluminum lactate injections. No significant correlations between the concentrations of aluminum and of TBARS were found in the whole brain. Subcellular analysis revealed that aluminum lactate injections led to a significant increase in the concentration of aluminum in the mitochondrial fraction but had no significant effect on the concentration of peroxides in any subcellular fraction. These results suggest that aluminum accumulation induced by the aluminum lactate administration associates with the acceleration of lipid peroxidation in rat brain. Furthermore, these data indicate that the pro-oxidant effect of aluminum may be indirect and concentration independent. The experimental conditions used here provide an animal model of aluminum accumulation in the brain that should prove useful for further investigations of the mechanisms of aluminum neurotoxicity.

Obata T. 2003. Phytic acid suppresses 1-methyl-4-phenylpyridinium ion-induced hydroxyl radical generation in rat striatum. Brain Res 978(1-2):241-244.
Abstract: The present study examined the antioxidant effect of phytic acid on iron (II)-enhanced hydroxyl radical (.OH) generation induced by 1-methyl-4-phenylpyridinium ion (MPP+) in the extracellular fluid of rat striatum. Rats were anesthetized, and sodium salicylate in Ringer's solution (0.5 nmol/mul/min) was infused through a microdialysis probe to detect the generation of .OH as reflected by the non-enzymatic formation of 2,3-dihydroxybenzoic acid (DHBA) in the striatum. Phytic acid (100 muM) did not significantly decrease the levels of MW-induced .OH formation trapped as 2,3-DHBA. To confirm the generation of .OH by the Fenton-type reaction, iron (II) was infused through a microdialysis probe. Introduction of iron (II) (10 muM) enhanced MPP+ induced .OH generation. However, phytic acid significantly suppressed iron (II)-enhanced .OH formation after MPP+ treatment (n=6, P<0.05). These results suggest that the antiradical effect of phytic acid occurs by chelating iron required for the MPP+-enhanced .OH generation via the Fenton-type reaction. (C) 2003 Elsevier Science B.V. All rights reserved.

Obata T. 2003. Calcium overload enhances hydroxyl radical generation by 1-methyl-4 phenylpyridinium ion (MPP+) in rat striatum. Brain Res 965(1-2): 287-289.
Abstract: We examined whether ouabain-induced Ca2+ overload increases hydroxyl radical ((OH)-O-.) generation by 1-methyl-4-phenylpyridinium ion (MPP+) in rat striatum. These elevations seem to induce lipid peroxidation

of striatum of rats, as detected by increases in non-enzymatic formation of 2,3-dihydroxybenzoic acid (DHBA) levels. Ouabain enhanced MPP+-induced (OH)-O-. formation trapped as DHBA. Moreover, when iron (II) was administered to MPP+ then ouabain (100 muM)-pretreated animals, a marked elevation in the level of DHBA was observed, as compared with the iron (II)-only-treated animals. These results suggests that Ca2+ overload might enhance (OH)-O-. generation by MPP+ in rat striatum. (C) 2002 Elsevier Science B.V. All rights reserved.

Nulton-Persson AC, Starke DW, Mieyal JJ, Szweda LI. 2003. Reversible inactivation of alpha-ketoglutarate dehydrogenase in response to alterations in the mitochondrial glutathione status. Biochemistry (Mosc) 42 (14):4235-4242.
Abstract: In a previous study, we found that treatment of rat heart mitochondria with H2O2 resulted in a decline and subsequent recovery in the rate of state 3 NADH-linked respiration. These effects were shown to be mediated by reversible alterations in NAD(P)H utilization and in the activities of specific Krebs cycle enzymes alpha-ketoglutarate dehydrogenase (KGDH) and succinate dehydrogenase. The purpose of the current study was to examine potential mechanism(s) by which H2O2 reversibly alters KGDH activity. We report here that inactivation is not simply due to direct interaction of H2O2 with KGDH. In addition, incubation of mitochondria with deferroxamine, an iron chelator, or 1,3-dimethyl-2-thiourea, an oxygen radical scavenger, prior to addition of H2O2 did not alter the rate or extent of inactivation. Thus, inactivation does not appear to involve a more potent oxygen radical formed upon metal-catalyzed oxidation. Inactive KGDH from H2O2-treated mitochondria was reactivated with dithiothreitol, implicating oxidation of a protein sulfhydryl(s). However, the thioredoxin system had no effect, indicating that enzyme inactivation is not due to the formation of intra- or intermolecular disulfide(s) or a sulfenic acid. Upon incubation of mitochondria with H2O2, reduced GSH levels fell rapidly prior to enzyme inactivation but recovered at the same time as enzyme activity. Importantly, treatment of inactive KGDH with glutaredoxin facilitated the GSH-dependent recovery of KGDH activity. Glutaredoxin is characterized as a specific and efficient catalyst of protein deglutathionylation. Thus, the results of the current study indicate that KGDH activity appears to be modulated through enzymatic glutathionylation and deglutathionylation. These studies demonstrate a novel mechanism by which KGDH activity and mitochondrial function can be modulated by redox status.

Norris EH, Giasson BI, Ischiropoulos H, Lee VMY. 2003. Effects of oxidative and nitrative challenges on alpha-synuclein fibrillogenesis involve distinct mechanisms of protein modifications. J Biol Chem 278(29):27230-27240.
Abstract: Filamentous inclusions of alpha-synuclein protein are hallmarks of neurodegenerative diseases collectively known as synucleinopathies. Previous studies have shown that exposure to oxidative and nitrative species stabilizes alpha-synuclein filaments in vitro, and this stabilization may be due to dityrosine cross-linking. To test this hypothesis, we mutated tyrosine residues to phenylalanine and generated recombinant wild type and mutant alpha-synuclein proteins. alpha-Synuclein proteins lacking some or all tyrosine residues form fibrils to the same extent as the wild type protein. Tyrosine residues are not required for protein cross-linking or filament stabilization resulting from transition metal-mediated oxidation, because higher M-r SDS-resistant oligomers and filaments stable to chaotropic agents are detected using all Tyr --> Phe alpha-synuclein mutants. By contrast, cross-linking resulting from exposure to nitrating agents required the presence of one or more tyrosine residues. Furthermore, tyrosine cross-linking is involved in filament stabilization, because nitrating agent-exposed assembled wild type, but not mutant alpha-synuclein lacking all tyrosine residues, was stable to chaotropic treatment. In addition, the formation of stable alpha-synuclein inclusions in intact cells after exposure to oxidizing and nitrating species requires tyrosine residues. These findings demonstrate that nitrative and/or oxidative stress results in distinct mechanisms of alpha-synuclein protein

modifications that can influence the formation of stable alpha-synuclein fibrils.

Nishida Y. 2003. Elucidation of endemic neurodegenerative diseases - a commentary. Zeitschrift Fur Naturforschung C-a Journal of Biosciences 58 (9-10): 752-758.
Abstract: Recent investigations of scrapie, Creutzfeldt-Jakob disease (CJD), and chronic wasting disease (CWD) clusters in Iceland, Slovakia and Colorado, respectively, have indicated that the soil in these regions is low in copper and higher in manganese, and it has been well-known that patients of ALS or Parkinson's disease were collectively found in the New Guinea and Papua islands, where the subterranean water (drinking water) contains much Al3+ and Mn2+ ions. Above facts suggest that these neurodegenerative diseases are closely related with the function of a metal ion. We have investigated the chemical functions of the metal ions in detail and established the unique mechanism of the oxygen activation by the transition metal ions such as iron and copper, and pointed out the notable difference in the mechanism among iron, aluminum and manganese ions. Based on these results, it has become apparent that the incorporation of Al (III) or Mn(II) in the cells induces the "iron-overload syndrome", which is mainly due to the difference in an oxygen activation mechanism between the iron ion and AI(III) or the Mn(II) ion. This syndrome highly promotes formation of hydrogen peroxide, and hydrogen peroxide thus produced can be a main factor to cause serious damages to DNA and proteins (oxidative stress), yielding a copper(II)- or manganese(II)-peptide complex and its peroxide adduct, which are the serious agents to induce the structural changes from the normal prion protein (PrPC) to abnormal disease-causing isoforms, PrPSc, or the formation of PrP 27-30 (abnormal cleavage at site 90 of the prion protein). It seems reasonable to consider that the essential origin for the transmissible spongiform encephalopathies (TSEs) should be the incorporation and accumulation of Al(III) and Mn(II) ions in the cells, and the sudden and explosive increase of scrapie and bovine spongiform encephalopathy (BSE) in the last decade may be partially due to "acid rain", because the acid rain makes Al(III) and Mn(II) ions soluble in the subterranean aquifers.

Miller K, Ochudlo S, Opala G, Smolicha W, Siuda J. 2003. [Parkinsonism in chronic occupational metallic mercury intoxication]. Neurol Neurochir Pol 37 Suppl 5:31-8.
Abstract: Parkinson syndrome occurs in the course of chemical intoxication, especially Mn, CS2, CO. It is rarely caused by chronic mercury intoxication. We present the case of 55 year old man who was exposed to metallic mercury vapor during 33 years of working in the chemical plant at the production of chlorine. On several occassions patient was removed from contact with Hg because of the symptoms of increased Hg absorption. At the age of 52 he developed hand tremor, balance and gait disturbance with bradykinesia, paresthesias of the upper extremities, neurobehavioral abnormalities, slight memory loss, and spatial disorientation. Psychoneurological examination revealed dementia, Parkinson's syndrome and ataxia of the lower limbs. Mercury excretion in the urine, which equaled 18.3 mu\g creatinine, confirmed exposure to Hg. MRI of the head revealed cortical and cerebellar atrophy. Electroneurography examination found features of subclinical peripheral sensory axonopathy of the upper limbs. Despite atypical clinical course (parkinsonismus) chronic mercury encephalopathy was diagnosed based on documented occupational exposure and diagnostic test results.

Mcdonnell L, Maginnis C, Lewis S, Pickering N, Antoniak M, Hubbard R, Lawson I, Britton J. 2003. Occupational exposure to solvents and metals and Parkinson's disease. Neurology 61(5):716-717.

Mandel S, Grunblatt E, Riederer P, Gerlach M, Levites Y, Youdim MBH. 2003. Neuroprotective strategies in Parkinson's disease - An update on progress. Cns Drugs 17(10):729-762.
Abstract: In spite of the extensive studies performed on postmortem

substantia nigra from Parkinson's disease patients, the aetiology of the disease has not yet been established. Nevertheless, these studies have demonstrated that, at the time of death, a cascade of events had been initiated that may contribute to the demise of the melanin-containing nigrostriatal dopamine neurons. These events include increased levels of iron and monoamine oxidase (MAO)-B activity, oxidative stress, inflammatory processes, glutamatergic excitotoxicity, nitric oxide synthesis, abnormal protein folding and aggregation, reduced expression of trophic factors, depletion of endogenous antioxidants such as reduced glutathione, and altered calcium homeostasis. To a large extent, the 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) and 6-hydroxydopamine (6-OHDA) animal models of Parkinson's disease confirm these findings. Furthermore,neuroprotection can be afforded in these models with iron chelators, radical scavenger antioxidants, MAO-B inhibitors, glutamate antagonists, nitric oxide synthase inhibitors, calcium channel antagonists and trophic factors. Despite the success obtained with animal models, clinical neuroprotection is much more difficult to accomplish. Although the negative studies obtained with the MAO-B inhibitor selegiline (deprenyl) and the antioxidant tocopherol (vitamin E) may have resulted from an inappropriate choice of drug (selegiline) or an inadequate dose (tocopherol), the niggling problem that still remains is why these drugs, and others, do work in animals while they fail in the clinic. One reason for this may be related to the fact that in normal human brains the number of dopaminergic neurons falls by around 3-5% every decade, while in Parkinson's disease this decline is greater. Brain autopsy studies have shown that by the time the disease is identified, some 70-75% of the dopamine-containing neurons have been lost. More sensitive reliable methods and clinical correlative markers are required to discern between confoundable symptomatic effects versus a possible neuroprotective action of drugs, namely, the ability to delay or forestall disease progression by protecting or rescuing the remaining dopamine neurons or even restoring those that have been lost. A number of other possibilities for the clinical failure of potential neuroprotectants also exist. First, the animal models of Parkinson's disease may not be totally reflective of the disease and, therefore, the chemical pathologies established in the animal models may not cause, or contribute to, the progression of the disease clinically. Second, because of the series of events occurring in neurodegeneration and our ignorance about which of these factors constitutes the primary event in the pathogenic process, a single drug may not be adequate to induce neuroprotection and, as a consequence, use of a cocktail of drugs may be more appropriate. The latter concept receives support from recent complementary DNA (cDNA) microarray gene expression studies, which show the existence of a gene cascade of events occurring in the nigrostriatal pathway of MPTP, 6-OHDA and methamphetamine animal models of Parkinson's disease.

Lorenzl S, Albers DS, Relkin N, Ngyuen T, Hilgenberg SL, Chirichigno J, Cudkowicz ME, Beal MF. 2003. Increased plasma levels of matrix metalloproteinase-9 in patients with Alzheimer's disease. Neurochem Int 43 (3):191-196.

Abstract: Matrix metalloproteinases (MMPs) may play a role in the pathophysiology of Alzheimer's disease (AD). MMP-9 and tissue inhibitors of metalloproteinases (TIMPs) are elevated in postmortem brain tissue of AD patients. MMPs and TIMPs are found in neurons, microglia, vascular endothelial cells and leukocytes. The aim of this study was to determine whether circulating levels of MMP-2, MMP-9, TIMP-1 and TIMP-2 are elevated in the plasma of AD patients. We compared AD patients to age- and gender-matched controls as well as to Parkinson's disease (PD) and amyotrophic lateral sclerosis (ALS) patients. There was constitutive expression of gelatinase A (MMP-2), and gelatinase B (MMP-9), in all the samples as shown by zymographic analysis. Levels of MMP-9 were significantly ($P = 0.003$) elevated in the plasma of AD patients as compared to controls. Plasma levels of MMP-2, TIMP-1 and TIMP-2 were unchanged. There were no significant changes of MMP-2, MMP-9, TIMP-1 and TIMP-2 levels in PD and ALS samples. TIMP-1 and TIMP-2 were

significantly correlated with MMP-9 in the AD patients. ApoE genotyping of plasma samples showed that levels of MMP-2, TIMP-1 and TIMP-2 and MMP-9 were not significantly different between the ApoE subgroups. These findings indicate that circulating levels of MMP-9 are increased in AD and may contribute to disease pathology. (C) 2003 Elsevier Science Ltd. All rights reserved.

Lorenzl S, Albers DS, Lewitt PA, Chirichigno JW, Hilgenberg SL, Cudkowicz ME, Beal MF. 2003. Tissue inhibitors of matrix metalloproteinases are elevated in cerebrospinal fluid of neurodegenerative diseases. J Neurol Sci 207 (1-2):71-76.
Abstract: Matrix metalloproteinases (MMPs) are implicated in the pathogenesis of diseases such as Alzheimer's Disease (AD) and amyotrophic lateral sclerosis (ALS). Increased expression of MMP-9 and TIMPs has been reported in postmortem AD and ALS brain tissue, as well as in ALS cerebrospinal fluid (CSF) and plasma. Although individual studies of MMP and TIMP expression in CSF have included AD and ALS samples, there are no studies comparing the expression of these proteins between neurodegenerative diseases. We measured the levels of matrix metalloproteinases (MMPs)-2 and -9 and the tissue inhibitor of MMPs (e.g. TIMP-1 and TIMP-2) in CSF samples from patients with Parkinson's Disease (PD), Huntington's Disease (HD), AD and ALS as compared to age-matched control patients. There was constitutive expression of the proform of gelatinase A (proMMP-2) on zymography gels in all CSF samples. Unexpectedly, there was an additional gelatinolytic band at 130 kDa of unknown etiology in the CSF samples of patients with PD (61% of patients studied), AD (61%), HD (25%) and ALS (39%). Levels of TIMP-1 were significantly elevated in CSF samples from all disease groups. TIMP-2 was significantly increased in CSF of AD and HD patients. MMP-2 levels did not differ significantly between groups. These findings show that TIMPs are elevated in the CSF of patients with neurodegenerative diseases suggesting a potential role of these endogenous inhibitors of matrix metalloproteinases in neurodegenerative diseases. (C) 2003 Elsevier Science B.V. All rights reserved.

Liu Y, Simon JD. 2003. Isolation and biophysical studies of natural eumelanins: Applications of imaging technologies and ultrafast spectroscopy. Pigment Cell Res 16(6):606-618.
Abstract: The major pigments found in the skin, hair, and eyes of humans and other animals are melanins. Despite significant research efforts, the current understanding of the molecular structure of melanins, the assembly of the pigment within its organelle, and the structural consequences of the association of melanins with protein and metal cations is limited. Likewise, a detailed understanding of the photochemical and photophysical properties of melanins has remained elusive. Many types of melanins have been studied to date, including natural and synthetic model pigments. Such studies are often contradictory and to some extent the diversity of systems studied may have detracted from the development of a basic understanding of the structure and function of the natural pigment. Advances in the understanding of the structure and function of melanins require careful characterization of the pigments examined so as to assure the data obtained may be relevant to the properties of the pigment in vivo. To address this issue, herein the influence of isolation procedures on the resulting structure of the pigment is examined. Sections describing the applications of new technologies to the study of melanins follow this. Advanced imaging technologies such as scanning probe microscopies are providing new insights into the morphology of the pigment assembly. Recent photochemical studies on photoreduction of cytochrome c by different mass fraction of sonicated natural melanins reveal that the photogeneration of reactive oxygen species (ROS) depends upon aggregation of melanin. Specifically, aggregation mitigates ROS photoproduction by UV-excitation, suggesting the integrity of melanosomes in tissue may play an important role in the balance between the photoprotective and photodamaging behaviors attributed to melanins. Ultrafast laser spectroscopy studies of melanins are providing insights into

the time scales and mechanisms by which melanin dissipates absorbed light energy.

Liu X, Lee B, Tjalkens R. 2003. Characterization of nigro-striatal deficits in a mouse model of manganese-induced parkinsonism. Toxicol Sci 72(1):21.

Liu JM, Hankinson SE, Stampfer MJ, Rifai N, Willett WC, Ma J. 2003. Body iron stores and their determinants in healthy postmenopausal US women. Am J Clin Nutr 78(6):1160-1167.
Abstract: Background: Data on the determinants of body iron stores in middle-aged women are sparse. Objective: We prospectively evaluated nondietary and dietary determinants of iron stores. Design: Using blood samples collected in 1989-1990, we measured plasma ferritin concentrations in 620 healthy postmenopausal women aged 44-69 y who participated in the Nurses' Health Study. Food-frequency questionnaires completed in 1980, 1984, and 1986 were used to calculate average dietary intakes. Generalized linear regression and multiple logistic regression models were used to assess the association between plasma ferritin and its determinants. Results: Among these postmenopausal women, the median plasma ferritin concentration was 73.8 ng/mL (interquartile range: 41.6 - 125.8 ng/mL), 2.7% were iron depleted (ferritin concentration < 12 ng/mL), and 9.8% had an elevated ferritin concentration (> 200 ng/mL). Age, time since menopause, time since the last postmenopausal hormone (PMH) use, body mass index, iron supplement use, and alcohol and heme-iron intakes were positively associated with ferritin concentrations, whereas PMH use, physical activity, aspirin use, and gastrointestinal ulcer were inversely related. The association between heme-iron intake and ferritin was most apparent among the women who consumed > 30 g alcohol/d. Conclusions: Our prospective data confirm that in postmenopausal women, intakes of heme iron, supplemental iron, and alcohol are dietary determinants of plasma ferritin, and age, PMH use, body mass index, physical activity, aspirin use, and gastrointestinal ulcer are nondietary determinants.

Lin MT, Beal MF. 2003. The oxidative damage theory of aging. Clinical Neuroscience Research 2(5-6):305-315.
Abstract: The oxidative stress theory of aging postulates that age-associated reductions in physiologic functions are caused by a slow steady accumulation of oxidative damage to macromolecules, which increases with age and which is associated with life expectancy of organisms. A corollary is that the rate of aging should be retarded by attenuation of oxidative damage. A large body of evidence has accumulated in support of this hypothesis. Increases in oxidative damage to DNA, proteins, and lipids have all been found with normal aging. Genetic manipulation of oxidative damage can increase life expectancy in animals. Overexpression of Cu/Zn superoxide dismutase or manganese superoxide dismutase appears to extend life span. Overexpression of methionine sulfoxide reductase in Drosophila resulted in a 70% increase in survival, and a 50% reduction in methionine sulfoxide reductase in mice resulted in a 30% reduction in life span. Caloric restriction, which extends life span, also reduces oxidative stress. Manipulation of gene expression in Drosophila with phenylbutyrate significantly increases lifespan, and is associated with a 50-fold increase in expression of manganese superoxide dismutase. We recently further examined the mitochondrial DNA theory of aging, which proposes that mitochondrial DNA accumulates mutations with age and that these contribute to the physiological decline in aging. Using a PCR-cloning-sequencing strategy, we found a significant increase in aggregate burden of mitochondrial DNA point mutations in brain in elderly subjects compared to younger subjects. The average aggregate mutational burden in elderly subjects was 2x10(-4) mutations per base. The bulk of these mutations were individually rare point mutations, and 60% changed an amino acid. Cytochrome oxidase activity correlated negatively with increased mutational burden. These findings bolster the possibility that oxidative damage to mitochondrial DNA may play a significant role in normal aging.

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Lin AMY, Fan SF, Yang DM, Hsu LL, Yang CHJ. 2003. Zinc-induced apoptosis in substantia nigra of rat brain: Neuroprotection by vitamin D3. Free Radic Biol Med 34(11):1416-1425.
Abstract: Accumulation of transition metals has been suggested to be responsible for the deteriorated nigrostriatal dopaminergic system in Parkinson's patients. In the present study, the mechanism underlying the zinc-induced neurotoxicity was investigated in the nigrostriatal dopaminergic system in vivo. Our 6-methoxy-8-paratoluene sulfonamide quinoline fluorescence study showed zinc translocation in the infused nigral cells after intranigral infusion of zinc. Furthermore, lipid peroxidation in the zinc-infused substantia nigra was consistently elevated 4 h to 7 d after the infusion. At the same time, an abrupt increase in cytosolic cytochrome c content in the infused substantia nigra was observed 4 h after zinc infusion and gradually decreased to basal levels 7 d after infusion. Both TUNEL-positive neurons and DNA fragmentation, indicatives of apoptosis, were detected in the zinc-infused substantia nigra. Furthermore, striatal dopamine content was reduced 7 d after the infusion. In attempt to prevent zinc-induced neurotoxicity, vitamin D3 was systemically administered. Zinc-induced increases in lipid peroxidation and cytosolic cytochrome c in the infused substantia nigra were prevented by this treatment. Moreover, zinc-induced reduction in striatal dopamine content was attenuated after vitamin D3 treatment. Our in vivo data suggest that zinc-induced oxidative stress may result in apoptosis followed by reduced dopaminergic function in the nigrostriatal dopaminergic system. Furthermore, vitamin D3 prevented zinc-induced oxidative injuries in the rat brain. (C) 2003 Elsevier Inc.

Li ZM, Jansen M, Pierre SR, Figueiredo-Pereira ME. 2003. Neurodegeneration: linking ubiquitin/proteasome pathway impairment with inflammation. International Journal of Biochemistry & Cell Biology 35(5):547-552.
Abstract: Neurodegenerative disorders have been reported to be associated with accumulation of ubiquitinated proteins in neuronal inclusions and also with signs of inflammation. In these disorders, the abnormal protein aggregates may, themselves, trigger the expression of inflammatory mediators, such as, cyclooxygenase 2 (COX-2). Impairment of the ubiquitin/proteasome pathway may contribute to this neurodegenerative process. Accordingly, proteasome inhibitors and oxidative stressors such as cadmium, were found to decrease survival, induce the accumulation of ubiquitinated proteins and elicit up-regulation of cyclooxygenase 2 in neuronal cell cultures. Products of cyclooxygenase 2, such as prostaglandin J2, can, in turn, increase the levels of ubiquitinated proteins and also cause cyclooxygenase 2 up-regulation, creating a "self-destructive" feedback mechanism. In neurodegenerative disorders characterized by neuronal inclusions containing ubiquitinated proteins, a disruption of the ubiquitin/proteasome pathway may, therefore, act in conjunction with cyclooxygenase 2 up-regulation to exacerbate the neurodegenerative process. Cyclooxygenase 2 inhibitors and agents that prevent protein aggregation could be of therapeutic value to these forms of neurodegeneration. (C) 2003 Elsevier Science Ltd. All rights reserved.

Levy BS, Nassetta WJ. 2003. Neurologic effects of manganese in humans: A review. Int J Occup Environ Health 9(2):153-163.
Abstract: Manganese, which enters the body primarily via inhalation, can damage the nervous system and respiratory tract, as well as have other adverse effects. Occupational exposures occur mainly in mining, alloy production, processing, ferro-manganese operations, welding, and work with agrochemicals. Among the neurologic effects is an irreversible parkinsonian-like syndrome. An estimated 500,000 to 1.5 million people in the United States have Parkinson's disease, and physicians need to consider manganese exposure in its differential diagnosis. Since 1837, there have been many reports of cases and case series describing manganese toxicity. More recently, there have been epidemiologic studies of its adverse effects on health. Occupational medicine physicians can play

critical roles in preventing the adverse health effects of manganese.

Levites Y, Amit T, Mandel S, Youdim MBH. 2003. Neuroprotection and neurorescue against A beta toxicity and PKC-dependent release of non-amyloidogenic soluble precursor protein by green tea polyphenol (-)-epigallocatechin-3-gallate. FASEB J 17(3):952-954.
Abstract: Green tea extract and its main polyphenol constituent (-)-epigallocatechin-3-gallate (EGCG) possess potent neuroprotective activity in cell culture and mice model of Parkinson's disease. The central hypothesis guiding this study is that EGCG may play an important role in amyloid precursor protein (APP) secretion and protection against toxicity induced by beta-amyloid (Abeta). The present study shows that EGCG enhances (similar to6-fold) the release of the non-amyloidogenic soluble form of the amyloid precursor protein (sAPPalpha) into the conditioned media of human SH-SY5Y neuroblastoma and rat pheochromocytoma PC12 cells. sAPP release was blocked by the hydroxamic acid-based metalloprotease inhibitor Ro31-9790, which indicated mediation via secretase activity. Inhibition of protein kinase C (PKC) with the inhibitor GF109203X, or by down-regulation of PKC, blocked the EGCG-induced sAPPalpha secretion, suggesting the involvement of PKC. Indeed, EGCG induced the phosphorylation of PKC, thus identifying a novel PKC-dependent mechanism of EGCG action by activation of the non-amyloidogenic pathway. EGCG is not only able to protect, but it can rescue PC12 cells against the beta-amyloid (Abeta) toxicity in a dose-dependent manner. In addition, administration of EGCG (2 mg/kg) to mice for 7 or 14 days significantly decreased membrane-bound holoprotein APP levels, with a concomitant increase in sAPPalpha levels in the hippocampus. Consistently, EGCG markedly increased PKCalpha and PKCepsilon in the membrane and the cytosolic fractions of mice hippocampus. Thus, EGCG has protective effects against Abeta-induced neurotoxicity and regulates secretory processing of non-amyloidogenic APP via PKC pathway.

Levenson CW. 2003. Iron and Parkinson's disease: Chelators to the rescue? Nutr Rev 61(9):311-313.
Abstract: The essential metal iron has long been implicated in the neuronal damage associated with Parkinson's disease. Recent findings show that iron chelation may prevent the reductions in dopamine and motor disturbances associated with this disease, and suggest the need to examine the role of dietary iron and the use of metal chelators in neurodegenerative disorders.

Lee EN, Lee SY, Lee D, Kim J, Paik SR. 2003. Lipid interaction of alpha-synuclein during the metal-catalyzed oxidation in the presence of Cu2+ and H2O2. J Neurochem 84(5):1128-1142.
Abstract: alpha-Synuclein co-exists with lipids in the Lewy bodies, a pathological hallmark of Parkinson's disease. Molecular interaction between alpha-synuclein and lipids has been examined by observing lipid-induced protein self-oligomerization in the presence of a chemical coupling reagent of N- (ethoxycarbonyl)-2-ethoxy-1,2-dihydroquinoline. Lipids such as phosphatidic acid, phosphatidylinositol, phosphatidylserine, phosphatidylethanolamine, and even arachidonic acid induced the self-oligomerization whereas phosphatidylcholine did not affect the protein. Because the oligomerizations occurred from critical micelle concentrations of the lipids, the self interaction of alpha-synuclein was shown to be a lipid-surface dependent phenomenon with head group specificity. By employing beta-synuclein and a C-terminally truncated alpha-synuclein (alpha-syn97), the head-group dependent self-oligomerization was demonstrated to occur preferentially at the N-terminal region while the fatty acid interaction leading to the protein self-association required the presence of the acidic C-terminus of alpha-synuclein. In the presence of Cu2+ and H2O2 , phosphatidylinositol (PI), along with other acidic lipids, actually enhanced the metal-catalyzed oxidative self-oligomerization of alpha-synuclein. The dityrosine crosslink formation responsible for the PI-enhanced covalent self-oligomerization was more sensitive to variation of copper concentrations than that of H2O2 during the metal-catalyzed oxidation. The

enhancement by PI was shown to be due to facilitation of copper localization to the protein because actual binding affinity between copper and alpha-synuclein increased from K-d of 44.7 mum to 5.9 mum in the presence of the lipid. Taken together, PI not only affects alpha-synuclein to be more self-interactive by providing the lipid surface, but also enhances the metal-catalyzed oxidative protein self-oligomerization by facilitating copper localization to the protein when the metal and H2O2 are provided. This observation therefore could be implicated in the formation of Lewy bodies as lipids and metal-catalyzed oxidative stress have been considered to be a part of pathological causes leading to the neurodegeneration.

Lee CS, Han ES, Lee WB. 2003. Antioxidant effect of phenelzine on MPP+-induced cell viability loss in differentiated PC12 cells. Neurochem Res 28(12): 1833-1841.
Abstract: Phenelzine, deprenyl, and antioxidants (SOD, catalase, ascorbate, or rutin) reduced the loss of cell viability in differentiated PC12 cells treated with 250 muM MPP+, whereas N-acetylcysteine and dithiothreitol did not inhibit cell death. Phenelzine reduced the condensation and fragmentation of nuclei caused by MPP+ in PC12 cells. Phenelzine and deprenyl prevented the MPP+-induced decrease in mitochondrial membrane potential, cytochrome c release, formation of reactive oxygen species, and depletion of GSH in PC12 cells. Phenelzine revealed a scavenging action on hydrogen peroxide and reduced the hydrogen peroxide-induced cell death in PC12 cells, whereas deprenyl did not depress the cytotoxic effect of hydrogen peroxide. Both compounds reduced the iron and EDTA-mediated degradation of 2-deoxy-D-ribose degradation. The results suggest that phenelzine attenuates the MPP+-induced viability loss in PC12 cells by reducing the alteration of mitochondrial membrane permeability that seems to be mediated by oxidative stress.

Lacreuse A, Herndon JG. 2003. Effects of estradiol and aging on fine manual performance in female rhesus monkeys. Horm Behav 43(3):359-366.
Abstract: Aging is characterized by a progressive deterioration of motor function related to dysfunctions of the nigrostriatal system. Because estrogen has been reported to protect dopaminergic neurons and to improve the motor deficits associated with Parkinson's disease, we hypothesized that it would partially reverse the age-related decline of motor function in normal aging. We tested the effects of estrogen treatment and withdrawal on fine motor performance in five aged (21-24 years old) and five young (6-9 years old) ovariectomized female rhesus monkeys. The tests required the monkeys to use each hand to retrieve a Life Saver candy from metal rods bent in shapes of different complexity. Monkeys were tested twice a week for 8 consecutive weeks, during treatment with placebo or ethinyl estradiol (EE2) in alternating 14-day blocks. Each behavioral test was videotaped and subsequently scored for the duration and the success of the first trial on each shape. Both groups of monkeys improved rapidly with practice in speed and success of retrieval. The older monkeys were slower but as successful as the young monkeys in retrieving the candy. The left hand was faster than the right hand for both the aged and young females. We failed to detect any effect of EE2 treatment on speed or success of retrieval in either group. These results confirm the slowing of fine motor performance with aging in female rhesus monkeys. They also indicate that estradiol, at least as administered in this study, does not benefit fine manual performance. (C) 2003 Elsevier Science (USA). All rights reserved.

Kurup RK, Kurup PA. 2003. Hypothalamic digoxin-mediated model for Parkinson's disease. Int J Neurosci 113(4):515-536.
Abstract: The isoprenoid pathway produces four key metabolites important in cellular function-digoxin (endogenous membrane Na+-K+ ATPase inhibitor), dolichol (important in N-glycosylation of proteins), ubiquinone (free-radical scavenger), and cholesterol (component of cellular membranes). This study assessed the changes in the isoprenoid pathway and the consequences of its dysfunction in Parkinson's disease (PD). There

was an elevation in plasma HMG CoA reductase activity, serum digoxin and dolichol levels, and a reduction in serum magnesium, RBC membrane Na+-K+ ATPase activity, and serum ubiquinone levels. Serum tryptophan, serotonin, strychnine, nicotine, and quinolinic acid were elevated, while tyrosine, morphine, dopamine, and noradrenaline were decreased. The total serum glycosaminoglycans (GAG) and glycosaminoglycan fractions (except chondroitin sulphates and hyaluronic acid), the activity of GAG degrading enzymes, carbohydrate residues of serum glycoproteins, the activity of glycohydrolase-beta galactosidase, and serum glycolipids were elevated. HDL cholesterol was reduced and free fatty acids increased. The RBC membrane glycosaminoglycans, hexose and fucose residues of glycoproteins and cholesterol were reduced, while phospholipid was increased. The activity of all serum free-radical scavenging enzymes, concentration of glutathione, alpha tocopherol, iron binding capacity, and ceruloplasmin decreased significantly in PD, while the concentration of serum lipid peroxidation products and nitric oxide increased. A dysfunctional isoprenoid pathway and related cascade are important in the pathogenesis of Parkinson's disease. A hypothalamic digoxin mediated model for Parkinson's disease is also postulated.

Kunikowska G, Jenner P. 2003. Alterations in m-RNA expression for Cu,Zn-superoxide dismutase and glutathione peroxidase in the basal ganglia of MPTP-treated marmosets and patients with Parkinson's disease. Brain Res 968(2):206-218.
Abstract: Alterations occurring in the antioxidant enzymes, copper, zinc-dependent superoxide dismutase (Cu,Zn-SOD) and glutathione peroxidase (GPX) following nigral dopaminergic denervation are unclear. We now report on the distribution and levels of m-RNA for Cu,Zn-SOD and GPX in basal ganglia of normal and 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP)-treated common marmosets, and in normal individuals and patients with Parkinson's disease (PD) using in situ hybridization histochemistry and oligodeoxynucleotide (single-stranded DNA) probes. Cu,Zn-SOD and GPX m-RNA was present throughout basal ganglia (nucleus accumbens, caudate-putamen, globus pallidus, substantia nigra) in the common marmoset, with the highest levels being in substantia nigra (SN). Following MPTP induced nigral cell loss, Cu,Zn-SOD m-RNA levels were decreased in all areas but the SNr, and particularly in SNc (71%, $P<0.001$). MPTP-treatment had no effect on GPX m-RNA expression in any area of basal ganglia. Cu,Zn-SOD and GPX m-RNA was also present in the normal human SN. In PD, however, Cu,Zn-SOD m-RNA was significantly decreased (89%, $P<0.005$) in SNc, and there was a near-complete loss of GPX m-RNA in both SNc (100%, $P<0.005$) and SNr (88%, $P<0.005$). The loss of Cu,Zn-SOD m-RNA in SNc in MPTP-treated marmosets and patients with PD suggests that it is primarily located in dopaminergic neuronal cell bodies. The loss of GPX m-RNA in SNc in PD also suggests a localisation to dopaminergic cell bodies, but the similar change in SNr may indicate its presence in dopaminergic neurites. In contrast, the absence of change in GPX m-RNA in MPTP-treated primates appears to rule out its presence in dopaminergic cells in this species, but this may only be apparent and may reflect increased expression in glial cells following acute toxin treatment.
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Kulich SM, Chu CT. 2003. Role of reactive oxygen species in extracellular signal-regulated protein kinase phosphorylation and 6-hydroxydopamine cytotoxicity. Journal of Biosciences 28(1):83-89.
Abstract: A number of reports indicate the potential for redox signalling via extracellular signal-regulated protein kinases (ERK) during neuronal injury. We have previously found that sustained ERK activation contributes to toxicity elicited by 6-hydroxydopamine (6-OHDA) in the B65 neuronal cell line. To determine whether reactive oxygen species (ROS) play a role in mediating ERK activation and 6-OHDA toxicity, we examined the effects of catalase, superoxide dismutase (SOD1), and metalloporphyrin antioxidants ('SOD mimetics') on 6-OHDA-treated cells. We found that catalase and metalloporphyrin antioxidants not only conferred protection against 6-OHDA but also inhibited development of sustained ERK phosphorylation in

both differentiated and undifferentiated B65 cells. However, exogenously added SOD1 and heat-inactivated catalase had no effect on either toxicity or sustained ERK phosphorylation. This correlation between antioxidant protection and inhibition of 6-OHDA-induced sustained ERK phosphorylation suggests that redox regulation of ERK signalling cascades may contribute to neuronal toxicity.

Kobayashi H, Uchida M, Sato I, Suzuki T, Hossain MM, Suzuki K. 2003. Neurotoxicity and brain regional distribution of manganese in mice. Journal of Toxicology-Toxin Reviews 22(4):679-689.
Abstract: Manganese (Mn) is known to cause neurotoxicity in the central nervous system similar to Parkinsonism in man, but the mechanism underlying remains to be well clarified. Catalepsy is used to observe Parkinsonism in laboratory animals. In the present study, effects of repeated injection of Mn chloride (MnCl2) on catalepsy, dopamine receptors and distribution of Mn-54 in the brain were investigated. [Methods] Female ICR mice were injected with 0, 10, 30 or 50 mg/kg/day of MnCl2 for 3 days, and examined for catalepsy and the binding ability of striatum, hippocampus and cerebral cortex to [H-3]haloperidol to detect and change of dopamine D-2-receptors. Whole-body burden and disposition of Mn-54 in the brain regions and liver were determined after the repeated injection of (MnCl2)-Mn-54. Mice were given L-dopa at 25 mg/kg 2 hr prior to MnCl2 injection to examined if the catalepsy was abolished. [Results] Mice showed catalepsy following injection of MnCl2 at 50 mg/kg, but not with less than 30 mg/kg. The catalepsy initiated about 60, 60 and 30 min after injection of MnCl2 on the 1st, 2nd and 3rd day, respectively, and lasted for about 60 min. L-dopa slightly reversed the catalepsy. The binding of [H-3]haloperidol in the three brain regions from mice treated with MnCl2 was lower than that from control. The concentration of Mn-54 in the striatum and remaining areas, including substantia nigra, was the highest in the brain regions examined. [Conclusion] Since L-dopa slightly alleviated catalepsy by MnCl2, and binding of [H-3]haloperidol was decreased in brain regions, MnCl2 might induce catalepsy by suppressing D-2 receptors in the striatum-substantia nigra.

Kita T, Wagner GC, Nakashima T. 2003. Current research on methamphetamine-induced neurotoxicity: Animal models of monoamine disruption. Journal of Pharmacological Sciences 92(3):178-195.
Abstract: Methamphetamine (METH)-induced neurotoxicity is characterized by a long-lasting depletion of striatal dopamine (DA) and serotonin as well as damage to striatal dopaminergic and serotonergic nerve terminals. Several hypotheses regarding the mechanism underlying METH-induced neurotoxicity have been proposed. In particular, it is thought that endogenous DA in the striatum may play an important role in mediating METH-induced neuronal damage. This hypothesis is based on the observation of free radical formation and oxidative stress produced by autooxidation of DA consequent to its displacement from synaptic vesicles to cytoplasm. In addition, METH-induced neurotoxicity may be linked to the glutamate and nitric oxide systems within the striatum. Moreover, using knockout mice lacking the DA transporter, the vesicular monoamine transporter 2, c-fos, or nitric oxide synthetase, it was determined that these factors may be connected in some way to METH-induced neurotoxicity. Finally a role for apoptosis in METH-induced neurotoxicity has also been established including evidence of protection of bcl-2, expression of p53 protein, and terminal deoxynucleotidyl transferase-mediated dUTP nick-end labeling (TUNEL), activity of caspase-3. The neuronal damage induced by METH may reflect neurological disorders such as autism and Parkinson's disease.

Kim NH, Kang JH. 2003. Oxidative modification of neurofilament-L by copper-catalyzed reaction. Journal of Biochemistry and Molecular Biology 36(5): 488-492.
Abstract: Neurofilament-L (NF-L) is a major element of neuronal cytoskeletons and known to be important for. neuronal survival in vivo. Since oxidative stress might play a critical role in the pathogenesis of

neurodegenerative diseases, we investigated the role of copper and peroxide in the modification of NF-L. When disassembled NF-L was incubated with copper ion and hydrogen peroxide, then the aggregation of protein was proportional to copper and hydrogen peroxide concentrations. Dityrosine crosslink formation was obtained in copper-mediated NF-L aggregates. The copper-mediated modification of NF-L was significantly inhibited by thiol antioxidants, N-acetylcysteine, glutathione, and thiourea. A thioflavin-T binding assay was performed to determine whether the copper/H2O2 system-induced in vitro aggregation of NF-L displays amyloid-like characteristics. The aggregate of NF-L displayed thioflavin T reactivity, which was reminiscent of amyloid. This study suggests that copper-mediated NF-L modification might be closely related to oxidative reactions which may play a critical role in neurodegenerative diseases.

Killilea DW, Atamna H, Liao C, Ames BN. 2003. Iron accumulation during cellular senescence in human fibroblasts in vitro. Antioxidants & Redox Signaling 5 (5):507-516.
Abstract: Iron accumulates as a function of age in several tissues in vivo and is associated with the pathology of numerous age-related diseases. The molecular basis of this change may be due to a loss of iron homeostasis at the cellular level. Therefore, changes in iron content in primary human fibroblast cells (IMR-90) were studied in vitro as a model of cellular senescence. Total iron content increased exponentially during cellular senescence, resulting in 10-fold higher levels of iron compared with young cells. Low-dose hydrogen peroxide (H2O2) induced early senescence in IMR-90s and concomitantly accelerated iron accumulation. Furthermore, senescence-related and H2O2-stimulated iron accumulation was attenuated by N-tert-butylhydroxylamine (NtBHA), a mitochondrial antioxidant that delays senescence in vitro. However, SV40-transformed, immortalized IMR-90s showed no time-dependent changes in metal content in culture or when treated with H2O2 and/or NtBHA. These data indicate that iron accumulation occurs during normal cellular senescence in vitro. This accumulation of iron may contribute to the increased oxidative stress and cellular dysfunction seen in senescent cells.

Khan FH, Sen T, Chakrabarti S. 2003. Dopamine oxidation products inhibit Na+, K +-ATPase activity in crude synaptosomal-mitochondrial fraction from rat brain. Free Radic Res 37(6):597-601.
Abstract: The diverse damaging effects of dopamine (DA) oxidation products on brain subcellular components including mitochondrial electron transport chain have been implicated in dopaminergic neuronal death in Parkinson's disease. It has been shown in this study that DA (50-200 muM) causes dose-dependent inhibition of Na+, K+-ATPase activity of rat brain crude synaptosomal-mitochondrial fraction during in vitro incubation up to 2 h. The enzyme inactivation is prevented by catalase and the metal-chelator (diethylenetriamine penta-acetic acid) but not by superoxide dismutase or hydroxyl-radical scavengers like mannitol and dimethylsulphoxide (DMSO). Further, reduced glutathione and cysteine, markedly prevent DA-mediated inactivation of Na+, K+-ATPase. Under similar conditions of incubation, DA (200 muM) leads to the formation of quinoprotein adducts (protein-cysteinyl catechol) with synaptosomal-mitochondrial proteins and the phenomenon is also prevented by glutathione (5 mM) or cysteine (5 mM). The available data imply that the inactivation of Na+, K+-ATPase in this system involves both H2O2 and metal ions. The reactive quinones by forming adducts with protein thiols also probably contribute to the process, since reduced glutathione and cysteine which scavenge quinones from the system protect Na+, K+-ATPase from DA-mediated damage. The inactivation of neuronal Na+, K+-ATPase by DA may give rise to various toxic sequelae with potential implications for dopaminergic cell death in Parkinson's disease.

Ke Y, Qian ZM. 2003. Iron misregulation in the brain: a primary cause of neurodegenerative disorders. Lancet Neurology 2(4):246-253.
Abstract: High iron concentrations in the brains of patients and the discovery of mutations in the genes associated with iron metabolism in the

brain suggest that iron misregulation in the brain plays a part in neuronal death in some neuro-degenerative disorders, such as Alzheimer's, Parkinson's, and Huntington's diseases and Hallervorden-Spatz syndrome. Iron misregulation in the brain may have genetic and non-genetic causes. The disrupted expression or function of proteins involved in iron metabolism increases the concentration of iron in the brain. Disturbances can happen at any of several stages in iron metabolism (including uptake and release, storage, intracellular metabolism, and regulation). Increased brain iron triggers a cascade of deleterious events that lead to neurodegeneration. An understanding of the process of iron reglation in the brain, the proteins important in this process, and the effects of iron misregulation could help to treat or prevent neurodegenerative disrders.

Kaya M, Kalayci R, Arican N, Kucuk M, Elmas I. 2003. Effect of aluminum on the blood-brain barrier permeability during nitric oxide-blockade-induced chronic hypertension in rats. Biol Trace Elem Res 92(3):221-230.
Abstract: We examined the effect of aluminum on the permeability of the blood-brain barrier (BBB) during nitric oxide-blockade-induced chronic hypertension in rats. Animals were given the inhibitor of nitric oxide synthase, L-NAME (N-omega-nitro-L-arginine methyl ester), for 4 wk to induce chronic hypertension. Two groups of rats were given an intraperitoneal injection of aluminum chloride. The integrity of the BBB was assessed by a quantitative measurement for Evans blue (EB) dye. The arterial blood pressure in L-NAME- and L-NAME plus aluminum-treated animals was significantly elevated from 115+/-2.8 and 110+/-1.7 mm Hg to 174+/-5.2 and 175+/-4.8 mm Hg, respectively ($p < 0.01$). The EB dye content in the brain regions of the rats in the L-NAME group was increased, but there was no statistical significance compared to the saline group. The extravasation of EB dye was significantly increased in the brain regions of the animals treated with aluminum compared to the rats treated with saline ($p < 0.05$). A significantly higher EB dye content in the brain regions was observed in the L-NAME plus aluminum group compared to L-NAME, aluminum, and saline groups ($p < 0.01$). These findings indicate that exposure to a high level of aluminum leads to an additional increase in BBB permeability where nitric oxide-blockade-induced chronic hypertension potentiates the effect of aluminum to enhance BBB permeability to EB dye.

Kawahara M, Kato-Negishi M, Hosoda R, Imamura L, Tsuda M, Kuroda Y. 2003. Brain-derived neurotrophic factor protects cultured rat hippocampal neurons from aluminum maltolate neurotoxicity. J Inorg Biochem 97(1): 124-131.
Abstract: Aluminum is environmentally abundant but not an essential trace element. Although there is increasing evidence suggesting the implication of aluminum in the pathogenesis of Alzheimer's disease, it is still controversial. We found and report here that aluminum maltolate, a stable and hydrophilic aluminum complex, causes death of primary cultured rat hippocampal. neurons in a time- and dose-dependent manner. Degenerated neurons were TUNEL-positive. Immunohistochemical detection of synapsin I and micrombule associated protein 2 revealed the synapse loss between neurons intoxicated by aluminum maltolate. To explore the mechanism underlying its neurotoxicity, we administered various pharmacological compounds prior to the application of aluminum maltolate, and found that brain-derived neurotrophic factor (BDNF) markedly attenuated the neurotoxicity. Furthermore, aluminum maltolate inhibited the elevation of intracellular calcium levels caused by BDNF. Our results suggest the involvement of BDNF in the molecular mechanism underlying neurotoxicity induced by aluminum maltolate. (C) 2003 Elsevier Inc. All rights reserved.

Kawahara M. 2003. Aluminum-induced conformational changes of beta-amyloid protein and the pathogenesis of Alzheimer's disease . Journal of Health Science 49(5):341-347.
Abstract: Aggregation and subsequent conformational change of Alzheimer's beta-amyloid protein (AbetaP) enhance its neurotoxicity.

Therefore, factors that inhibit or promote conformational changes of AbetaP play crucial roles in the pathogenesis of Alzheimer's disease (AD). Moreover, recent studies have suggested that a common mechanism is based on the diverse diseases termed "conformational diseases" including neurodegenerative diseases such as AD, prion diseases, Parkinson's disease, and Huntington's disease. These diseases share similarity in the formation of beta-sheet containing amyloid fibrils by disease-related proteins and the introduction of apoptotic degeneration. Aluminum, an environmental risk factor for AD, is a widely used cross-linker that causes conformational changes of AbetaP and other proteins. This report reviews and discusses characteristics of aluminum-induced conformational changes of AbetaP and their implication in pathogenesis of AD. Taking together our results and those of numerous other studies, we hypothesize that aluminum-induced conformational changes enhance the neurotoxicity of AbetaP and lead to development of AD.

Kaur D, Yantiri F, Rajagopalan S, Kumar J, Mo JO, Boonplueang R, Viswanath V, Jacobs R, Yang L, Beal MF, Dimonte D, Volitaskis I, Ellerby L, Cherny RA, Bush AI, Andersen JK. 2003. Genetic or pharmacological iron chelation prevents MPTP-induced neurotoxicity in vivo: A novel therapy for Parkinson's disease. Neuron 37(6):899-909.
Abstract: Studies on postmortem brains from Parkinson's patients reveal elevated iron in the substantia nigra (SN). Selective cell death in this brain region is associated with oxidative stress, which may be exacerbated by the presence of excess iron. Whether iron plays a causative role in cell death, however, is controversial. Here, we explore the effects of iron chelation via either transgenic expression of the iron binding protein ferritin or oral administration of the bioavailable metal chelator clioquinol (CO) on susceptibility to the Parkinson's-inducing agent 1 -methyl-4-phenyl-1,2,3,6-tetrapyridine (MPTP). Reduction in reactive iron by either genetic or pharmacological means was found to be well tolerated in animals in our studies and to result in protection against the toxin, suggesting that iron chelation may be an effective therapy for prevention and treatment of the disease.

Kang JH, Kim KS. 2003. Enhanced oligomerization of the alpha-synuclein mutant by the Cu,Zn-superoxide dismutase and hydrogen peroxide system. Mol Cells 15(1):87-93.
Abstract: The alpha-synuclein is a major component of Lewy bodies that are found in the brains of patients with Parkinson's disease (PD). Also, two point mutations in this protein, A53T and A30P, are associated with rare familial forms of the disease. We investigated whether there are differences in the Cu,Zn-SOD and hydrogen peroxide system mediated-protein modification between the wild-type and mutant alpha-synucleins. When alpha-synuclein was incubated with both Cu,Zn-SOD and H2O2, then the amount of A53T mutant oligomerization increased relative to that of the wild-type protein. This process was inhibited by radical scavenger, spin-trapping agent, and copper chelator. These results suggest that the oligomerization of alpha-synuclein is mediated by the generation of the hydroxyl radical through the metal-catalyzed reaction. The dityrosine formation of the A53T mutant protein was enhanced relative to that of the wild-type protein. Antioxidant molecules, carnosine, and anserine effectively inhibited the wild-type and mutant proteins' oligomerization. Therefore, these compounds may be explored as potential therapeutic agents for PD patients. The present experiments, in part, may provide an explanation for the association between PD and the alpha-synuclein mutant.

Kamatani T, Yamamoto T, Yoneda K, Osaki T. 2003. Polymorphic mutations of the Mn-SOD gene in intact human lymphocytes and oral squamous cell carcinoma cell lines. Biochemistry and Cell Biology-Biochimie Et Biologie Cellulaire 81(1):43-50.
Abstract: Mutations of the superoxide dismutase (SOD) genes are associated with neoplastic and non-neoplastic diseases. However, the existence of polymorphic mutations of manganese SOD (Mn-SOD) has not

been explored in squamous cell carcinoma (SCC) cells or in normal cells. In the present study, we examined mutations in the 5' flanking region of the Mn-SOD gene and Mn-SOD mRNA using 10 human oral SCC (OSC) cell lines and intact lymphocytes obtained from 10 healthy donors and one patient with OSC. The polymerase chain reaction products of DNA obtained from lymphocytes revealed insertions at many sites (-1833, -1575, -1093, -1056, -325, -318, and -310) in 10 of the 11 donors. Transitions and (or) transversions were also observed at -1638 and -216 in lymphocytes from six donors and one donor, respectively. In DNA obtained from OSC cells, insertions and transitions and (or) transversions were more frequent than those in DNA from lymphocytes. In addition, deletions at -1341 and -1288 were observed in all lines except for one line. In these mutations, the transcription factor binding sites were not involved except for the AP-2 binding site (-102) in three cell lines. In Mn-SOD mRNA, Val at -9 position was varied to Ala in lymphocytes from two donors and three OSC cell lines, respectively. In the remaining cell lines, Mn-SOD mRNA from lymphocytes and OSC cell lines revealed heterozygosity (Ala/Val) and homozygosity (Val/Val), respectively. The Mn-SOD activities in lymphocytes were 3.8-5.8 x 10(-4) U/10(6) cells and the activities in OSC cell lines were 1.8-8.3 x 10 (-4) U/10(6) cells. These Mn-SOD activities were not correlated with the mutations of DNA and mRNA. From these results, it is indicated that polymorphic mutations of Mn-SOD exist in human normal cells and that the deletions might be obtained in the course of malignant transformation of OSC although decrease in Mn-SOD activity is not involved in the transformation.

Kalivendi SV, Kotamraju S, Cunningham S, Shang TS, Hillard CJ, Kalyanaraman B. 2003. 1-Methyl-4-phenylpyridinium (MPP+)-induced apoptosis and mitochondrial oxidant generation: role of transferrin-receptor-dependent iron and hydrogen peroxide. Biochem J 371:151-164.
Abstract: 1-Methyl-4-phenylpyridinium (MPP+) is a neurotoxin used in cellular models of Parkinson's Disease. Although intracellular iron plays a crucial role in MPP+-induced apoptosis, the molecular signalling mechanisms linking iron, reactive oxygen species (ROS) and apoptosis are still unknown. We investigated these aspects using cerebellar granule neurons (CGNs) and human SH-SY5Y neuroblastoma cells. MPP+ enhanced caspase 3 activity after 24 h with significant increases as early as 12 h after treatment of cells. Pre-treatment of CGNs and neuroblastoma cells with the metal] oporphyrin antioxidant enzyme mimic, Fe(III)tetrakis(4-benzoic acid)porphyrin (FeTBAP), completely prevented the MPP+-induced caspase 3 activity as did overexpression of glutathione peroxidase (GPxl) and pre-treatment with a lipophilic, cell-permeable iron chelator [N,N'-bis (2-hydroxybenzyl)ethylenediamine-N,N'-diacetic acid, HBED]. MPP+ treatment increased the number of TUNEL (terminal deoxynucleotidyl transferase-mediated dUTP nick-end-labelling)positive cells which was completely blocked by pre-treatment with FeTBAP. MPP+ treatment significantly decreased the aconitase and mitochondrial complex I activities; pre-treatment with FeTBAP, HBED and GPx1 overexpression reversed this effect. MPPI treatment increased the intracellular oxidative stress by 2-3-fold, as determined by oxidation of dichlorodihydrofluorescein and dihydroethidium (hydroethidine). These effects were reversed by pre-treatment of cells with FeTBAP and HBED and by GPx1 overexpression. MPP+-treatment enhanced the cell-surface transferrin receptor (TfR) expression, suggesting a role for TfR-induced iron uptake in MPP+ toxicity. Treatment of cells with anti-TfR antibody (IgA class) inhibited MPP+-induced caspase activation. Inhibition of nitric oxide synthase activity did not affect caspase 3 activity, apoptotic cell death or ROS generation by MPP+. Overall, these results suggest that MPP+-induced cell death in CGNs and neuroblastoma cells proceeds via apoptosis and involves mitochondrial release of ROS and TfR-dependent iron.

Jover R, Company L, Gutierrez A, Zapater P, Perez-Serra J, Girona E, Aparicio JR, Perez-Mateo M. 2003. Minimal hepatic encephalopathy and extrapyramidal signs in patients with cirrhosis. Am J Gastroenterol 98(7): 1599-1604.

Abstract: OBJECTIVES: Two types of neurological dysfunction can occur in compensated cirrhosis: 1) extrapyramidal signs related to the accumulation of manganese in the basal ganglia and 2) milder degrees of cognitive impairment known as minimal hepatic encephalopathy (mHE). We assessed whether there was any relationship between both disorders in 42 patients with compensated cirrhosis. METHODS: Minimal hepatic encephalopathy was diagnosed using a battery of manual neuropsychological tests. Cognitive functioning was assessed by the Mini-Mental State Examination. Extrapyramidal signs were evaluated by the Columbia scale. RESULTS: Minimal hepatic encephalopathy was diagnosed in 15 (35.7%) patients. A total of 52.4% of patients showed significant extrapyramidal signs. Scores for the Columbia scale were hiaher in the presence of mHE (mean +/- SD, 16.0 10.9 vs 5.3 +/- 7. 1, p = 0.0004). In the bivariate analysis, mHE, Child-Pugh score, and Mini-Mental State Examination score were significantly associated with extrapyramidal signs. whereas in the multivariate analysis, mHE was the only independent variable related to extrapyramidal signs. CONCLUSIONS: There was a link between extrapyramidal signs and diagnosis of mHE based on manual neuropsychological testing. This finding may be explained by the influence of extrapyramidal manifestations on test performance or by a real pathophysiological relationship between both disorders. Further studies are necessary to resolve this question. (C) 2003 by Am. Coll. of Gastroenterology.

Jellinger KA. 2003. Neuropathological spectrum of synucleinopathies. Mov Disord 18:S2-S12.
Abstract: Synucleinopathies comprise a diverse group of neurodegenerative proteinopathies that share common pathological lesions composed of aggregates of conformational and posttranslational modifications of alpha-synuclein in selected populations of neurons and glia. Abnormal filamentous aggregates of misfolded alpha-synuclein protein are the major components of Lewy bodies, dystrophic (Lewy) neurites, and the Papp-Lantos filaments in oligodendroglia and neurons in multiple system atrophy linked to degeneration of affected brain regions. The synucleinopathies include (1) Lewy body disorders and dementia with Lewy bodies, (2) multiple system atrophy (MSA), and (3) Hallervorden-Spatz disease. (1) The pathological diagnosis of Lewy body disorders and dementia with Lewy bodies is established by validated consensus criteria based on semiquantitative assessment of subcortical and cortical Lewy bodies as their common hallmarks. They are accompanied by subcortical multisystem degeneration with neuronal loss and gliosis with or without Alzheimer pathologic state. Lewy bodies also occur in numerous other disorders, including pure autonomic failure, neuroaxonal dystrophies, and various amyloidoses and tauopathies. (2) Multiple system atrophy, a sporadic, adult-onset degenerative movement disorder of unknown cause, is characterized by alpha-synuclein-positive glial cytoplasmic and rare neuronal inclusions throughout the central nervous system associated with striatonigral degeneration, olivopontocerebellar atrophy, and involvement of medullar and spinal autonomic nuclei. (3) In neurodegeneration with brain iron accumulation type I, or Hallervorden-Spatz disease, alpha-synuclein is present in axonal spheroids and glial and neuronal inclusions. While the identity of the major components of Lewy bodies suggests that a pathway leading from normal soluble to abnormal misfolded filamentous proteins is central for their pathogenesis, regardless of the primary disorder, there are conformational differences in alpha-synuclein between neuronal and glial aggregates, showing nonuniform mapping for its epitopes. Despite several cellular and transgenic models, it is not clear whether inclusion body formation is an adaptive/neuroprotective or a pathogenic reaction/process generated in response to different, mostly undetermined, functional triggers linked to neurodegeneration. (C) 2003 Movement Disorder Society.

Huang CC, Weng YH, Lu CS, Chu NS, Yen TC. 2003. Dopamine transporter binding in chronic manganese intoxication. J Neurol 250(11):1335-1339.
Abstract: Chronic exposure to manganese may induce parkinsonism

similar to idiopathic Parkinson's disease (PD). However, clinical manifestations of manganism also have some features different from PD. The mechanisms of manganese-induced parkinsonism remain not fully understood. Tc-99m-TRODAT-1 is a cocaine analogue that can bind to the dopamine transporter (DAT) site reflecting the function of presynaptic dopaminergic terminals. The purpose of this study was to evaluate DAT function using Tc-99m-TRODAT-1 to investigate the integrity of the presynaptic dopaminergic terminals in manganese-induced parkinsonism. Brain Tc-99m-TRODAT-1 single photon emission computed tomography was performed in 4 patients with chronic manganese intoxication in a ferromanganese smelting plant in Taiwan. Twelve PD patients and 12 healthy volunteers served as abnormal and normal controls, respectively. Clinically, all manganism patients had a bradykinetic-rigid syndrome. The scores of the Unified Parkinson's Disease Rating Scale ranged between 19 and 64. The uptake values of the Tc-99m-TRODAT-1 were 0.868+/-0.136 in the right corpus striatum and 0.865+/-0.118 in the left, as compared with 0.951+/-0.059 and 0.956+/-0.058, respectively for the normal controls. The data were significantly higher than 0.250+/-0.070 and 0.317 +/-0.066 respectively for the PD patients. Interestingly, there was a mild decrease in the uptake of Tc-99m-TRODAT-1 in the putamen and the ratio of putamen and caudate when compared with the normal controls. Although the DAT shows a slight decrease in the putamen of manganism patients as compared with that of the normal controls, the data indicate that the presynaptic dopaminergic terminals are not the main target of chronic manganese intoxication. In addition Tc-99m-TRODAT-1 SPECT can provide a useful, convenient and inexpensive tool for differentiation between chronic manganism and PD.

Henkin RI, Hoetker JD. 2003. Deficient dietary intake of vitamin E in patients with taste and smell dysfunctions: Is vitamin E a cofactor in taste bud and olfactory epithelium apoptosis and in stem cell maturation-and development? Nutrition 19(11-12):1013-1021.
Abstract: OBJECTIVES: We reviewed dietary intake of several nutrients in a large group of patients with taste and smell dysfunction, compared intake of these nutrients with standard values, and recognized that intake of vitamin E was significantly less than that of most other nutrients. Based on this observation we attempted to develop an hypothesis of the possible role vitamin E might play in these sensory disorders. METHODS: Vitamin E intake was measured in 250 patients with taste,and smell dysfunctions. RESULTS: Intake of the vitamin was 3.2 +/- 0.2 mg/d (mean +/- standard error of,the mean), or 36 +/- 2% of the recommended daily allowance, an intake significantly below that considered adequate. This diminished intake occurred with normal intake of total calories protein; fat; carbohydrate; several vitamins, including thiamin, niacin, and pyridoxine; and the trace metals zinc, copper, and iron. CONCLUSIONS: Although specific relations between vitamin E intake and smell and taste dysfunctions are unclear, the non-antioxidant roles of vitamin E indicate that it is a factor in apoptosis, cellular signaling, and growth of various cell lines, suggesting that this vitamin may play a role in growth and development of stem cells in taste buds and olfactory epithelium. (C) Elsevier Inc. 2003.

He Y, Thong PS, Lee T, Leong SK, Mao BY, Dong F, Watt F. 2003. Dopaminergic cell death precedes iron elevation in MPTP-injected monkeys. Free Radic Biol Med 35(5):540-547.
Abstract: Though increasing lines of evidence suggest that iron accumulation and iron-induced oxidative stress might be important pathological factors responsible for substantia nigra (SN) cell death in Parkinson's disease (PD), it is still unknown whether iron accumulation is a primary cause or consequence of nigral cell death. Using nuclear microscopy, iron histochemistry, TUNEL method for apoptosis detection, and tyrosine hydroxylase (TH) immumohistochemistry, the present study investigated possible changes in iron contents in the SN and correlations of dopaminergic cell death progression with the process of iron accumulation in 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine(MPTP)-induced parkinsonian monkey from 1 d to 18 months after MPTP administration.

Our study demonstrated that apoptosis occurred in the ipsilateral SN at 1 d after MPTP injection and the number of TH-positive cells decreased significantly from 1 week onward. However, iron content was significantly increased in the ipsilateral SN from 4.5 months to 18 months after MPTP injection, and the iron increase was significantly correlated to the extent of dopaminergic cell death. These results suggest that dopaminergic cell death induced by MPTP administration might lead to iron accumulation in the monkey SN, and increased iron might contribute to the progression of nigral degeneration. (C) 2003 Elsevier Inc.

Hayflick SJ, Westaway SK, Levinson B, Zhou B, Johnson MA, Ching KHL, Gitschier J. 2003 . Genetic, clinical, and radiographic delineation of Hallervorden-Spatz syndrome. N Engl J Med 348(1):33-40.
Abstract: Background: Hallervorden-Spatz syndrome is an autosomal recessive disorder characterized by dystonia, parkinsonism, and iron accumulation in the brain. Many patients with this disease have mutations in the gene encoding pantothenate kinase 2 (PANK2); these patients are said to have pantothenate kinase-associated neurodegeneration. In this study, we compared the clinical and radiographic features of patients with Hallervorden-Spatz syndrome with and without mutations in PANK2. Methods: One hundred twenty-three patients from 98 families with a diagnosis of Hallervorden-Spatz syndrome were classified on the basis of clinical assessment as having classic disease (characterized by early onset with rapid progression) or atypical disease (later onset with slow progression). Their genomic DNA was sequenced for PANK2 mutations. Results: All patients with classic Hallervorden-Spatz syndrome and one third of those with atypical disease had PANK2 mutations. Whereas almost all mutations in patients with atypical disease led to amino acid changes, those in patients with classic disease more often resulted in predicted protein truncation. Patients with atypical disease who had PANK2 mutations were more likely to have prominent speech-related and psychiatric symptoms than patients with classic disease or mutation-negative patients with atypical disease. In all patients with pantothenate kinase-associated neurodegeneration, whether classic or atypical, T(sub 2)-weighted magnetic resonance imaging (MRI) of the brain showed a specific pattern of hyperintensity within the hypointense medial globus pallidus. This pattern was not seen in any patients without mutations. Conclusions: PANK2 mutations are associated with all cases of classic Hallervorden-Spatz syndrome and one third of cases of atypical disease. A specific MRI pattern distinguishes patients with PANK2 mutations. Predicted levels of pantothenate kinase 2 protein correlate with the severity of disease.

Hautot D, Pankhurst QA, Khan N, Dobson J. 2003. Preliminary evaluation of nanoscale biogenic magnetite in Alzheimer's disease brain tissue. Proc R Soc Lond B Biol Sci 270:S62-S64.
Abstract: Elevated iron levels are associated with many types of neurodegenerative disease, such as Alzheimer's, Parkinson's and Huntington's diseases. However, these elevated iron levels do not necessarily correlate with elevated levels of the iron storage or transport proteins, ferritin and transferrin. As such, little is known about the form of this excess iron. It has recently been proposed that some of the excess iron in neurodegenerative tissue may be in the form of the magnetic iron oxide magnetite (Fe3O4). We demonstrate, for the first time to our knowledge, using highly sensitive superconducting quantum interference device (SQUID) magnetometry, that the concentrations of magnetite are found to be significantly higher in three samples of Alzheimer's disease tissue than in three age- and sex-matched controls. These results have implications, not only for disease progression, but also for possible early diagnosis.

Hashimoto T, Elder CM, Okun MS, Patrick SK, Vitek JL. 2003. Stimulation of the subthalamic nucleus changes the firing pattern of pallidal neurons. J Neurosci 23(5):1916-1923.
Abstract: To clarify the mechanism underlying improvement of parkinsonian signs by high-frequency electrical stimulation (HFS) of the

subthalamic nucleus (STN), we investigated the effects of STN HFS on neuronal activity of the internal and external segment of the globus pallidus (GPi and GPe, respectively) in two rhesus monkeys rendered parkinsonian by administration of 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine. A scaled-down version of the chronic stimulating electrode used in humans, consisting of four metal contacts 0.50 mm in length each separated by 0.50 mm, was implanted through a cephalic chamber targeting the STN. Histological reconstruction revealed that the cathode was located in the STN in both monkeys. Extracellular recordings from a total of 110 pallidal neurons during STN stimulation were performed. Poststimulus time histograms of single neurons triggered by 2 Hz STN stimulation pulses at 2.4-3.0 V revealed short-latency excitations at 2.5-4.5 and 5.5-7.0 msec after stimulation onset and inhibitions at 1.0-2.5, 4.5-5.5, and 7.0-9.0 msec for both GPe and GPi neurons. These short-latency responses were present with 136 Hz stimulation, at voltages effective for alleviation of parkinsonian signs, resulting in a significant increase in mean discharge rate and a stimulus-synchronized regular firing pattern. These results indicate that activation of the STN efferent fibers and resultant changes in the temporal firing pattern of neurons in GPe and GPi underlie the beneficial effect of HFS in the STN in Parkinson's disease and further support the role of temporal firing patterns in the basal ganglia in the development of Parkinson's disease and other movement disorders.

Hashimoto M, Rockenstein E, Masliah E. 2003. Transgenic Models of Alpha-Synuclein Pathology - Past, Present, and FutureVolume 991. p 171-188. Parkinson's Disease: the Life Cycle of the Dopamine Neuron: Annals of the New York Academy of Sciences.
Abstract: Accumulation and toxic conversion to protofibrils of alpha-synuclein has been associated with neurological disorders such as Parkinson's disease (PD), Lewy body disease, multiple system atrophy, neurodegeneration with brain iron accumulation type 1, and Alzheimer's disease. In recent years, modeling these disorders in transgenic (tg) mice and flies has helped improve understanding of the pathogenesis of these diseases and has established the basis for the development of new experimental treatments. Overexpression of alpha-synuclein in tg mice in a region- and cell-specific manner results in degeneration of selective circuitries accompanied by motor deficits and inclusion formation similar to what is found in PD and related disorders. Furthermore, studies in singly and doubly tg mice have shown that toxic conversion and accumulation can be accelerated by alpha-synuclein mutations associated with familial parkinsonism, by amyloid beta peptide 1-42 (Abeta 1-42), and by oxidative stress. In contrast, molecular chaperones such as Hsp70 and close homologues such as alpha-synuclein have been shown to suppress toxicity. Similar studies are underway to evaluate the effects of other modifying genes that might play a role in alpha-synuclein ubiquitination. Among them considerable interest has been placed on the role of molecules associated with familial parkinsonism (Parkin, UCHL-1). Furthermore, studying the targeted overexpression of alpha-synuclein and other modifier genes in the nigrostriatal and limbic system by using regulatable promoters, lentiviral vectors, and siRNA will help improve understanding of the molecular mechanisms involved in selective neuronal vulnerability, and it will aid the development of new treatments.

Harris ED. 2003. Basic and clinical aspects of copper. Crit Rev Clin Lab Sci 40(5): 547-586.
Abstract: An oxygen-rich atmosphere obligated living organisms to cope with reactive oxygen species (O-2(-), H2O2, OH.) that were the unavoidable by-products of cellular metabolism. As a redox cofactor Cu was selected as a co-catalyst for numerous biological processes, many involving the utilization of oxygen. Inadequate or excessive intake of Cu can be pathogenic and life-threatening. Mutations to genes that code for Cu-transporting ATPase enzymes are the molecular basis of Wilson and Menkes diseases and more recently Cu has been identified as a preemptory factor-in amyloid and prion diseases.-This review is dedicated to bringing historical and timely information on Cu transport, metabolism

and homeostasis to the attention of those not familiar with this important mineral. Other comprehensive reviews are available to the interested readers.(1-4)

Greener M. 2003. Steeling for a possible iron-Parkinson connection. Scientist 17 (11):34.

Gerlach M, Double KL, Ben-Shachar D, Zecca L, Youdim MBH, Riederer P. 2003. Neuromelanin and its interaction with iron as a potential risk factor for dopaminergic neurodegeneration underlying Parkinson's disease. Neurotoxicity Research 5(1-2):35-43.
Abstract: Neuromelanin (NM) is a granular, dark brown pigment produced in some but not all of the dopaminergic neurons of the human substantia nigra (SN). In Parkinson's disease (PD) the pigmented dopaminergic neurons of the SN degenerate, suggesting that this process is related to the presence of NM. As yet it is unknown whether NM in the parkinsonian brain differs from that found in healthy tissue and thus may fulfil a different role within this tissue. The function of NM within the pigmented neurons is unknown but other melanins are believed to play a protective role via attenuation of free radical damage. Experimental evidence suggests that NM may also exhibit this characteristic, possibly by direct inactivation of free radical species or via its ability to chelate transition metals, such as iron. NM has the ability to bind a variety of metals, seven per cent of isolated NM is reported to consist of Fe, Cu, Zn and Cr. Iron is of particular interest as this metal is highly concentrated within the SN. Up to 20 per cent of the total iron contained in the SN from normal subjects is bound within NM. Further, it was demonstrated that NM contains a protein component and that iron is bound to NM in the ferric form. Increased tissue iron found in the parkinsonian SN may saturate iron-chelating sites on NM, and a looser association between iron and NM may result in an increased, rather than decreased, production of free radical species. It is hypothesized that this redox-active iron could be released and involved in a Fenton-like reaction leading to an increased production of oxidative radicals. The resultant radical-mediated cytotoxicity may contribute to cellular damage observed in PD.

Gellein K, Garruto RM, Syversen T, Sjobakk TE, Flaten TP. 2003. Concentrations of Cd, Co, Cu, Fe, Mn, Rb, V, and Zn in formalin-fixed brain tissue in amyotrophic lateral sclerosis and parkinsonism-dementia complex of Guam determined by high-resolution ICP-MS. Biol Trace Elem Res 96(1-3):39-60.
Abstract: Amyotrophic lateral sclerosis (ALS) and parkinsonism-dementia complex (PDC) are neurodegenerative disorders that occurred with extremely high frequency among the native population on Guam, especially in the 1950s and 1960s, but have substantially declined over the last half-century. The etiology of these diseases is unknown, but the most plausible hypothesis centers on imbalances in essential and toxic metals. We have determined the concentrations of Cd, Co, Cu, Fe, Mn, Rb, V, and Zn in formalin-fixed brain tissue collected during the period 1979-1983 from eight Guamanian patients with ALS, four with PDC, and five control subjects using high-resolution inductively coupled plasma-mass spectrometry. The concentrations of Cd are markedly and significantly elevated both in gray and white matter in ALS, but not in PDC patients. The concentrations of Zn are elevated for both patient groups, in both gray and white matter, but only the difference in gray matter for PDC is significant. For the other metals, no significant differences are found.

Fredriksson A, Schroder N, Archer T. 2003. Neurobehavioural deficits following postnatal iron overload: I spontaneous motor activity. Neurotoxicity Research 5(1-2):53-76.
Abstract: Five experiments that induced postnatal iron overload in mice are described. In Experiment 1, exposure of NMRI mice to different doses (iron succinate: 0.0, 3.7 or 37.0 mg Fe2+/kg b. w., p.o.) on postnatal days (PD) 10 - PD12 indicated marked disruptions of spontaneous motor behaviour and habituation in the 37.0 mg Fe2+/kg dose group, and to a lesser extent the 3.7 mg Fe2+/kg dose group. Analysis of brain iron content indicated

significantly more total iron (mug/g) in the basal ganglia, but not frontal cortex of the higher, 37 mg/kg, dose group. In Experiment 11, newborn NMRI mice were administered Fe2+ (7.5 mg/kg, b.w.) at either PD 3-5, PD 10-12 or PD 19-21, or vehicle (saline). Marked deficits in spontaneous motor behaviour and habituation were obtained in the mice administered iron during PD 10-12, and to a much lesser extent at PD 3-5. Analysis of total brain iron content indicated significantly more iron (mg/g) in the basal ganglia, but not frontal cortex of mice from PD 3-5 and PD 10-12 Fe2+ treatment groups. In Experiment 111, the interactive effects of postnatal iron overload and administration of 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) to adult C57 BL/6 mice were examined by postnatal administration of iron (Fe2+) 7.5 mg/kg, b. wt., p.o. or vehicle (saline) at PD 10-12 followed, at 3-months of age, by administration of either MPTP (2 x 20 or 2 x 40 mg/kg, s.c.) or saline. Postnatal Fe2+ (7.5 mg/kg) caused drastic disruptions of spontaneous motor activity and habituation at behavioural testing (4-months age), and adult MPTP treatment potentiated these disruptions. Neurochemical deficits in dopamine (DA) and its metabolites (dihydroxyphenylacetic acid, DOPAC; homovanillic acid, HVA) induced by MPTP treatment were exacerbated by prior postnatal administration of Fe2+. The analysis of total iron content (mg/g) in brain regions indicated notably elevated levels in the basal ganglia, but not in the frontal cortex of mice administered Fe2+ at PD 10-12. MPTP-treated mice displayed severe depletions of DA, DOPAC and HVA in both the striatal and frontal cortical regions, i.e. Veh-MPTP40 as well as Fe-MPTP20 and Fe-MPTP40 groups, compared to the saline-treated (Vehicle) mice at 4-months of age, with lesser depletions by the Veh-MPTP20 group. In Experiment IV it was indicated that postnatal iron induced marked deficits (hypoactivity), initially, in all three parameters of motor activity at the 5.0 and 7.5 mg/kg doses, and to a lesser extent at the 2.5 mg/kg dose. Later combination with MPTP (2 x 40 mg/kg) potentiated severely these deficits. During the final period of testing a marked hyperactivity was obtained for the two higher dose groups; this effect was abolished in mice administered MPTP. In Experiment V, postnatal iron-induced deficits were alleviated in a dose-related manner by co-administration of the uncompetitive glutamate receptor antagonist, dizolcipine (MK-801), with a subthreshold dose of L-Dopa. Iron-overload during the immediate postnatal period seems detrimental for several aspects of functional and neurobiological. development.

Fredriksson A, Archer T. 2003. Effect of postnatal iron administration on MPTP-induced behavioral deficits and neurotoxicity: behavioral enhancement by L-Dopa-MK-801 co-administration. Behav Brain Res 139(1-2):31-46. Abstract: Two experiments were performed to investigate the interactive effects of postnatal iron administration and adult MPTP treatment upon the function of C57 B1/6 mice tested at adult age and to ascertain the possible ameliatory effects of a subthreshold dose of L-Dopa co-administered with different doses of the uncompetitive glutamate antagonist, MK-801. Experiment I indicated that postnatal iron induced marked deficits (hypoactivity), initially, in all three parameters of motor activity at the 5.0 and 7.5 mg/kg doses, and to a lesser extent at the 2.5 mg/kg dose. Later combination with MPTP (2 x 40 mg/kg) potentiated severely these deficits. During the final period of testing a marked hyperactivity was obtained for the two higher dose groups; this effect was abolished in mice administered MPTP. Experiment 11 indicated that the deficits in motor activity parameters induced by postnatal iron at 7.5 mg/kg were alleviated in a dose-related manner by the co-administration of the uncompetitive glutamate antagonist, MK-801, with a subthreshold dose of L-Dopa. Postnatal iron (7.5 mg/kg) administration followed by low doses of MPTP (2 x 20 mg/kg) 3 months later virtually abolished all motor activity. The combination of these compounds increased also the motor activity of mice treated with MPTP (2 x 20 mg/kg) or mice treated with the combination of postnatal iron and MPTP. The combination of MK-801 with L-Dopa increased locomotor (0.3 mg/kg), rearing (0.1 and 0.3 mg/kg) and total activity (0.3 mg/kg) of iron-treated mice during the initial, hypoactive 30-min period of testing. Locomotor activity (0.1 mg/kg) of MPTP-treated mice was

increased too during this period. During the final 30-min period of testing all three parameters of activity (locomotion, 0.3 mg/kg; rearing and total activity, 0.1 and 0.3 mg/kg) were enhanced in the iron-treated mice, locomotion (0.1 mg/kg) and rearing (0.1 mg/kg) in the iron plus MPTP treated mice and only locomotion (0.1 mg/kg) in the MPTP-treated mice. In control mice (vehicle + saline), the higher doses of MK-801 (0.1 and 0.3 mg/kg) enhanced both locomotor and total activity. Analyses of total iron concentration in the frontal cortex and basal ganglia of Fe2+ and vehicle treated mice indicated that marked elevations basal ganglia iron levels of the 5.0 and 7.5 mg/kg groups, later injected either saline or MPTP, were obtained (Experiment 1). In Experiment 11, iron concentrations in the basal ganglia were elevated in both the Fe2+-sal and Fe2+-MPTP groups to 170 and 177% of Veh.-sal values, respectively. There was a significant increase in the frontal cortex of iron-treated mice later administered either saline or MPTP (2 x 40 mg/kg) in Experiment I as well as in those given iron followed by MPTP (2 x 20 mg/kg) in Experiment II. The implications of iron overload in parkinsonism seem confirmed by the interactive effects of postnatal administration of the metal followed by adult MPTP treatment upon motor activity and the activity-enhancing effects of co-administration of L-Dopa with MK-801. (C) 2002 Elsevier Science B.V. All rights reserved.

Fonck C, Baudry M. 2003. Rapid reduction of ATP synthesis and lack of free radical formation by MPP+ in rat brain synaptosomes and mitochondria. Brain Res 975(1-2):214-221.
Abstract: MPTP is a neurotoxin thought to damage dopaminergic neurons through free radical formation. MPTP is metabolized in the brain to MPP+, which is taken up into dopaminergic neurons via the doparnme transporter and assumed to impair mitochondrial function. We used striatal synaptosomes and telencephalic mitochondria to further investigate MPP+ mechanism of action. For comparison, the respiratory toxins FCCP, a cyanide analog that uncouples mitochondrial ATP production, and rotenone, a NADH dehydrogenase inhibitor, were also tested. FCCP, MPP+ and rotenone caused a rapid but stable decrease in [H-3]dopamine (DA) uptake by striatal synaptosomes. Two free radical scavengers, the salen-manganese complex EUK-134, and the spin trap s-PBN, did not prevent MPP+-induced decrease in DA uptake. However, addition of ATP during synaptosome preparation resulted in partial recovery of MPP+-induced [H-3]DA uptake decrease. Generation of oxygen free radicals by treatment of telencephalic mitochondria with MPP+, FCCP, or rotenone, was evaluated by measuring DCF fluorescence, while light emission by the luciferin-luciferase complex was used to determine ATP levels. MPP+, unlike rotenone, did not produce oxygen free radicals, but rather blocked ATP production in mitochondria, as did FCCP and rotenone. Taken together, these results suggest that MPP+ toxicity, at least during its initial stages, is primarily due to a decrease in ATP synthesis by mitochondria and not to free radical formation. (C) 2003 Elsevier Science B.V. All rights reserved.

Ferretti G, Bacchetti T, Moroni C, Vignini A, Curatola G. 2003. Copper-induced oxidative damage on astrocytes: protective effect exerted by human high density lipoproteins. Biochimica Et Biophysica Acta-Molecular and Cell Biology of Lipids 1635(1):48-54.
Abstract: In the present study, we confirmed that copper ions induce oxidative damage in human astrocytes in culture, as demonstrated by the significant increase in the levels of hydroperoxides and in the fluorescence intensity of the fluorescent probe dichloro-dihydrofluorescein diacetate (H (2)DCFDA). The compositional changes were associated with a significant decrease in cell viability in astrocytes treated with 10 muM Cu+ + with respect to control cells. Astrocytes incubated with copper ions in the presence of high density lipoproteins (HDL) isolated from plasma of normolipermc subjects showed lower levels of hydroperoxides and a higher cell viability with respect to cells oxidized alone. Moreover, a significant decrease in the levels of hydroperoxides was observed in oxidized astrocytes treated with HDL. These results demonstrate that HDL exert a protective role against lipid peroxidation. The protective effect could be related to the ability of HDL to bind metal ions at the lipoprotein

surface and/or to a stimulation of the efflux of lipid hydroperoxides from cell membranes as demonstrated in other cell types. Oxidative damage of astrocytes was induced at a copper concentration similar to that observed in cerebrospinal fluid (CSF) of patients affected by neurodegenerative diseases such as Alzheimer's (AD) and Parkinson's diseases (PD). Lipoprotein particles similar for density and chemical composition to plasma HDL were recently isolated in human CSF, therefore, the protective role exerted by HDL against Cu++ - induced oxidative damage of astrocytes could be of physiological relevance. (C) 2003 Elsevier B.V. All rights reserved.

Felletschin B, Bauer P, Walter U, Behnke S, Spiegel J, Csoti I, Sommer U, Zeiler B, Becker G, Riess O, Berg D. 2003. Screening for mutations of the ferritin light and heavy genes in Parkinson's disease patients with hyperechogenicity of the substantia nigra. Neurosci Lett 352(1):53-56.
Abstract: Recently, an insertional mutation in the ferritin-L gene was reported in some patients with familial basal ganglia degeneration, which, however, could not be detected in another Parkinson's disease (PD) population. We investigated 186 PD patients, in whom an increased amount of iron of the substantia nigra (SN) was priorly identified by transcranial ultrasound, for mutations of the whole coding region of ferritin-L and ferritin-H by denaturing high-pressure liquid chromatography and subsequent sequencing. In the ferritin-L gene two silent mutations were detected. In the ferritin-H gene the sequence variation 161 A --> G was found in one patient but none of the 186 controls. Although functional analysis will show, whether this sequence variation might be causative for single cases of PD, the results indicate that mutations in the ferritin genes are not a common cause for PD with increased levels of iron of the SN. (C) 2003 Elsevier Ireland Ltd. All rights reserved.

Faucheux BA, Martin ME, Beaumont C, Hauw JJ, Agid Y, Hirsch EC. 2003. Neuromelanin associated redox-active iron is increased in the substantia nigra of patients with Parkinson's disease. J Neurochem 86(5):1142-1148.
Abstract: Degeneration of dopaminergic neurones during Parkinson's disease is most extensive in the subpopulation of melanized-neurones located in the substantia nigra pars compacta. Neuromelanin is a dark pigment produced in the dopaminergic neurones of the human substantia nigra and has the ability to bind a variety of metal ions, especially iron. Post-mortem analyses of the human brain have established that oxidative stress and iron content are enhanced in association with neuronal death. As redox-active iron (free Fe2+ form) and other transition metals have the ability to generate highly reactive hydroxyl radicals by a catalytic process, we investigated the redox activity of neuromelanin (NM)-aggregates in a group of parkinsonian patients, who presented a statistically significant reduction (- 70%) in the number of melanized-neurones and an increased non-heme (Fe3+) iron content as compared with a group of matched-control subjects. The level of redox activity detected in neuromelanin-aggregates was significantly increased (+ 69%) in parkinsonian patients and was highest in patients with the most severe neuronal loss. This change was not observed in tissue in the immediate vicinity of melanized-neurones. A possible consequence of an overloading of neuromelanin with redox-active elements is an increased contribution to oxidative stress and intraneuronal damage in patients with Parkinson's disease.

Egana JT, Zambrano C, Nunez MT, Gonzalez-Billault C, Maccioni RB. 2003. Iron-induced oxidative stress modify tau phosphorylation patterns in hippocampal cell cultures. Biometals 16(1):215-223.
Abstract: Oxidative stress phenomena have been related with the onset of neurodegenerative diseases. Particularly in Alzheimer Disease (AD), oxygen reactive species (ROS) and its derivatives can be found in brain samples of postmortem AD patients. However, the mechanisms by which oxygen reactive species can alter neuronal function are still not elucidated. There is a growing amount of evidence pointing to a role for mitochondrial damage as the source of free radicals involved in oxidative stress. Among the species that participate in the production of oxygen reactive radicals,

transition metals are one of the most important. Several reports have implicated the involvement of redox-active metals with the onset of different neurodegenerative diseases such as Alzheimer's Disease (AD), Progressive Supranuclear Palsy (PSP), Amyotrophic Lateral Sclerosis (ALS) and Parkinson's Disease (PD). On the other hand, our previous studies have indicated that Abeta-induced deregulation of the protein kinase Cdk5 associated with tau protein hyperphosphorylation constitute a critical pathway toward neurodegeneration. In the current paper we have shown that iron induces an imbalance in the function of Cdk5/p25 system of hippocampal neurons, resulting in a marked decrease in tau phosphorylation at the typical Alzheimer's epitopes. The loss of phosphorylated tau epitopes correlated with an increase in 4-hydroxy-nonenal (HNE) adducts revealing damage by oxidative stress. This effects on tau phosphorylation patterns seems to be a consequence of a decrease in the Cdk5/p25 complex activity that appears to result from a depletion of the activator p25, a mechanism in which calcium transients could be implicated.

Ebadi M, Sharma SK. 2003. Peroxynitrite and mitochondrial dysfunction in the pathogenesis of Parkinson's disease. Antioxidants & Redox Signaling 5(3): 319-335.
Abstract: Nitric oxide (NO), in excess, behaves as a cytotoxic substance mediating the pathological processes that cause neurodegeneration. The NO-induced dopaminergic cell loss causing Parkinson's disease (PD) has been postulated to include the following: an inhibition of cytochrome oxidase, ribonucleotide reductase, mitochondrial complexes 1, 11, and IV in the respiratory chain, superoxide dismutase, glyceraldehyde-3-phosphate dehydrogenase; activation or initiation of DNA strand breakage, poly(ADP-ribose) synthase, lipid peroxidation, and protein oxidation; release of iron; and increased generation of toxic radicals such as hydroxyl radicals and peroxynitrite. NO is formed by the conversion of L-arginine to L-citrulline by NO synthase (NOS). At least three NOS isoforms have been identified by molecular cloning and biochemical studies: a neuronal NOS or type I NOS (nNOS), an immunologic NOS or type 2 NOS (iNOS), and an endothelial NOS or type 3 NOS (eNOS). The enzymatic activities of eNOS or nNOS are induced by phosphorylation triggered by Ca2+ entering cells and binding to calmodulin. In contrast, the regulation of iNOS seems to depend on de novo synthesis of the enzyme in response to a variety of cytokines, such as interferon-gamma and lipopolysaccharide. The evidence that NO is associated with neurotoxic processes underlying PD comes from studies using experimental models of this disease NOS inhibitors can prevent 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP)-induced dopaminergic neurotoxicity. Furthermore, NO fosters dopamine depletion, and the said neurotoxicity is averted by nNOS inhibitors such as 7-nitroindazole working on tyrosine hydroxylase-immunoreactive neurons in substantia nigra pars compacta. Moreover, mutant mice lacking the nNOS gene are more resistant to MPTP neurotoxicity when compared with wild-type littermates. Selegiline, an irreversible inhibitor of monoamine oxidase B, is used in PD as a dopaminergic function-enhancing substance. Selegiline and its metabolite, desmethylselegiline, reduce apoptosis by altering the expression of a number of genes, for instance, superoxide dismutase, Bcl-2, Bcl-xl, NOS, c-Jun, and nicotinamide adenine nucleotide dehydrogenase. The selegiline-induced antiapoptotic activity is associated with prevention of a progressive reduction of mitochondrial membrane potential in preapoptotic neurons. As apoptosis is critical to the progression of neurodegenerative disease, including PD, selegiline or selegiline-like compounds to be discovered in the future may be efficacious in treating PD.

Double KL, Halliday GM, Henderson J, Griffiths FM, Heinemann T, Riederer P, Gerlach M. 2003. The doparnine receptor agonist lisuride attenuates iron-mediated dopaminergic neurodegeneration. Exp Neurol 184(1):530-535.
Abstract: Many dopamine agonists used in the treatment of Parkinson's disease are suggested to be potentially neuroprotective. On the basis of its structure, the dopamine agonist lisuride may share this characteristic. In

the current study discrete asymptomatic lesions were produced by the injection of iron-laden neuromelanin into the rat substantia nigra and the animals treated with lisuride to determine the protective potential of this substance. Two treatment regimes were utilised. In the neuroprotective protocol, animals were treated with 0.1 mg(.)kg(-1) lisuride twice daily 3 days prior to, and 7 days following, the iron lesion. In the neurorescue protocol, the animals received 0.1 mg(.)kg(-1) lisuride twice daily for 1 week beginning on the fourth day post surgery. Eight weeks post surgery, tyrosine hydroxylase-positive neurons surrounding the injection site (33% of total nigral volume) were counted. Dopamine neuron number in iron-lesioned animals was reduced to 50% of that in vehicle-injected animals. The absence of motoric disturbances or a striatal dopamine deficit in these animals suggests a subclinical dopaminergic lesion. Dopamine neuron number in the quantified area in sham-injected animals receiving lisuride or iron-lesioned animals receiving lisuride in both the neuroprotection and neurorescue groups were not significantly reduced. These results suggest that lisuride can protect neurons against iron-induced cell death and might thus be neuroprotective in Parkinson's disease. (C) 2003 Elsevier Inc. All rights reserved.

Double KL, Gerlach M, Schunemann V, Trautwein AX, Zecca L, Gallorini M, Youdim MBH, Riederer P, Ben-Shachar D. 2003. Iron-binding characteristics of neuromelanin of the human substantia nigra. Biochem Pharmacol 66(3):489-494.
Abstract: The vulnerability of the dopaminergic neurons of the substantia nigra (SN) in Parkinson's disease has been related to the presence of the pigment neuromelanin (NM) in these neurons. It is hypothesised that NM may act as an endogenous storage molecule for iron, an interaction suggested to influence free radical production. The current study quantified and characterised the interaction between NM and iron. Iron-binding studies demonstrated that both NM and synthetically-produced dopamine melanin contain equivalent numbers of high and low-affinity binding sites for iron but that the affinity of NM for iron is higher than that of synthetic melanin. Quantification of the total iron content in iron-loaded NM and synthetic melanin demonstrated that the iron-binding capacity of NM is 10-fold greater than that of the model melanin. This data was in agreement with the larger iron cluster size demonstrated by Mossbauer spectroscopy in the native pigment compared with the synthetic melanin. These findings are consistent with the hypothesis that NM may act as an endogenous iron-binding molecule in dopaminergic neurons of the SN in the human brain. The interaction between NM and iron has implications for disorders such as Parkinson's disease where an increase in iron in the SN is associated with increased indices of oxidative stress. (C) 2003 Elsevier Science Inc. All rights reserved.

Di JW, Bi SP. 2003. Effect of aluminum (III) on the conversion of dopachrome in the melanin synthesis pathway. Spectrochim Acta A Mol Biomol Spectrosc 59(8):1689-1696.
Abstract: The effect of aluminum ions on the kinetics and mode of the conversion of dopachrome (DC) in acidic environment has been studied using UV-Vis spectrophotometric and cyclic voltammetric methods. The DC conversion step is an important reaction in melanogenesis. Aluminum ions catalyze greatly the decarboxylative transformation of DC to give 5,6-dihydroxyindole (DHI) rather than 5,6-dihydroxyindole-2-carboxylic acid (DHICA) at pH 5.5, which enhance the ratio of formation DHI/DHICA in melanin synthesis pathway. The kinetics of DC conversion catalyzed by aluminum ions is dependent on the concentration of DC and aluminum ions. These results provide evidence that aluminum ions could play a role in the synthesis of melanin pathway in acidic condition through catalyzing the DC decarboxylative transformation to yield DHI and influence the melanin structure and properties. (C) 2002 Elsevier Science B.V. All rights reserved.

Di JW, Bi SP. 2003. Aluminum ions accelerated the oxidative stress of copper-mediated melanin formation. Spectrochim Acta A Mol Biomol Spectrosc 59

(13):3075-3083.
Abstract: A comparison between the effects of aluminum and cupric ions on the dopachrome (DC) conversion and the cooperation effect of the both ions in the DOPA oxidation to melanin pathway has been studied by UV-Vis spectrophotometric method. Both aluminum and cupric ions catalyze the DC conversion reaction, which is an important step in the melanin synthesis pathway. However, cupric ions catalyze the conversion of DC to yield 5,6-dihydroxyindole-2-carboxylic acid (DHICA) but the product of DC conversion catalyzed by aluminum is 5,6-dihydroxyindole (DHI). DOPA oxidation catalyzed by aluminum and cupric ions is studied in the presence of hydrogen peroxide. The results from our experiments provide evidence that aluminum can markedly increase the oxidative stress of copper-mediated the melanin formation and influence the properties of the melanin by means of changing the ratio of DHICA/DHI in the acidic environment (pH 5.5). (C) 2003 Elsevier Science B.V. All rights reserved.

Dev KK, Hofele K, Barbieri S, Buchman VL, Van Der Putten H. 2003. Part II: alpha-synuclein and its molecular pathophysiological role in neurodegenerative disease. Neuropharmacology 45(1):14-44.
Abstract: alpha-Synuclein (alphaSN) brain pathology is a conspicuous feature of several neurodegenerative diseases. These include prevalent conditions such as Parkinson's disease (PD), dementia with Lewy bodies (DLB), and the Lewy body variant of Alzheimer's disease (LBVAD), as well as rarer conditions including multiple systems atrophy (MSA), and neurodegeneration with brain iron accumulation type-1 (NBIA-1). Common in these diseases, some referred to as alpha-synucleinopathies, are microscopic proteinaceous insoluble inclusions in neurons and glia that are composed largely of fibrillar aggregates of alphaSN. This molecular form of alphaSN contrasts sharply with normal alphaSN, which is an abundant soluble presynaptic protein in brain neurons. alphaSN is a highly conserved protein in vertebrates and only seven of its 140 amino acids differ between human and mouse. Flies lack an aSN gene. Implicated in neurotoxicity are two alphaSN mutants (A53T and A30P) that cause extremely rare familial forms of PD, alphaSN fibrils and protofibrils, soluble protein complexes of alphaSN with 14-3-3 protein, and phosphorylated, nitrosylated, and ubiquitylated alphaSN species. Unlike rare forms of fPD caused by mutations in alphaSN, disease mechanisms in most alpha-synucleinopathies implicate wildtype alphaSN and seem to converge around oxidative damage and impairments in protein catabolism. It is not known whether these causalities involve alphaSN from the beginning, but defects in the handling of this protein seem to contribute to disease progression because accumulation of toxic alphaSN forms damage neurons. Here, we summarize the main structural features of alphaSN and its functions, and discuss the molecular alphaSN species implicated in human disease and transgenic animal models of alpha-synucleinopathy in fly and rodents. (C) 2003 Elsevier Science Ltd. All rights reserved.

Dekker MCJ, Giesbergen PC, Njajou OT, Van Swieten JC, Hofman A, Breteler MMB, Van Duijn CM. 2003. Mutations in the hemochromatosis gene (HFE), Parkinson's disease and parkinsonism. Neurosci Lett 348(2):117-119.
Abstract: Iron overload increases oxidative stress and may lead to neurodegenerative disease like Parkinson's disease (PD). We studied the role of mutations in the hemochromatosis gene HFE in PD and other parkinsonism (non-PD PS) in two population-based series. The first series consisted of 137 patients with PD and 47 with non-PD PS, and the second of 60 patients with PD and 25 with non-PD PS. In the first series, PD patients were significantly more often homozygous for the C282Y mutation than controls (P = 0.03). Patients with non-PD PS in both series were more often carriers for the C282Y mutation than controls (P = 0.009, P = 0.006, respectively). Our data are hampered by small numbers, yet suggest that the C282Y mutation increases the risk of PD and non-PD PS. The rarity of this genotype requires a large series of patients to prove our hypothesis. (C) 2003 Elsevier Science Ireland Ltd. All rights reserved.

Datla KP, Murray HE, Pillai AV, Gillies GE, Dexter DT. 2003. Differences in

dopaminergic neuroprotective effects of estrogen during estrous cycle. Neuroreport 14(1):47-50.
Abstract: Previous studies suggest that estrogen treatment protects nigrostriatal dopaminergic neurons, but have not examined whether the changes in estrogen levels during estrous cycle can influence the susceptibility of these neurons to neurotoxins. Here we show that the loss of dopaminergic neurons in the substantia nigra was greater in animals lesioned at diestrus (low estrogen) using 6-hydroxydopamine or buffered iron chloride, when compared with animals lesioned at proestrus (high estrogen). Lesioning at diestrus with 6-hydroxydopamine reduced the striatal dopamine content, whereas the dopamine content was preserved in animals lesioned at proestrus. The density of the dopamine transporter, upon which 6-hydroxydopamine toxicity is dependant, was lower when circulating estrogen was high. These results thus support a neuroprotectory role for estrogen.

Coimbra CG, Junqueira VBC. 2003. High doses of riboflavin and the elimination of dietary red meat promote the recovery of some motor functions in Parkinson's disease patients. Braz J Med Biol Res 36(10):1409-1417.
Abstract: Abnormal riboflavin status in the absence of a dietary deficiency was detected in 31 consecutive outpatients with Parkinson's disease (PD), while the classical determinants of homocysteine levels (B6, folic and B12) were usually within normal limits. In contrast, only 3 of acid, 10 consecutive outpatients with dementia without previous stroke had abnormal riboflavin status. The data for 12 patients who did not complete 6 months of therapy or did not comply with the proposed treatment paradigm were excluded from analysis. Nineteen PD patients (8 males and 11 females, mean age +/- SD = 66.2 +/- 8.6 years; 3, 3, 2, 5, and 6 patients in Hoehn and Yahr stages I to V) received riboflavin orally (30 mg every 8 h) plus their usual symptomatic medications and all red meat was eliminated from their diet. After 1 month the riboflavin status of the patients was normalized from 106.4 +/- 34.9 to 179.2 +/- 23 ng/ml (N = 9). Motor capacity was measured by a modification of the scoring system of Hoehn and Yahr, which reports motor capacity as percent. All 19 patients who completed 6 months of treatment showed improved motor capacity during the first three months and most reached a plateau while 5/19 continued to improve in the 3- to 6-month interval. Their average motor capacity increased from 44 to 71% after 6 months, increasing significantly every month compared with their own pretreatment status ($P < 0.001$, Wilcoxon signed rank test). Discontinuation of riboflavin for several days did not impair motor capacity and yellowish urine was the only side effect observed. The data show that the proposed treatment improves the clinical condition of PD patients. Riboflavin-sensitive mechanisms involved in PD may include glutathione depletion, cumulative mitochondrial DNA mutations, disturbed mitochondrial protein complexes, and abnormal iron metabolism. More studies are required to identify the mechanisms involved.

Chen KB, Lin AMY, Chiu TH. 2003. Systemic Vitamin D3 Attenuated Oxidative Injuries in the Locus Coeruleus of Rat BrainVolume 993. p 313-324. Neuroprotective Agents: Annals of the New York Academy of Sciences.
Abstract: Iron-induced oxidative injuries in locus coeruleus (LC), a major source of noradrenergic projections in the central nervous system (CNS), were investigated in chloral-hydrate anesthetized rats. Local infusion of iron dose-dependently elevated lipid peroxidation of iron-infused LC seven days after infusion. At the same time, norepinephrine content in the hippocampus ipsilateral to the iron-infused LC was decreased in a concentration-dependent manner. Our immunostaining study demonstrated reduced tyrosine hydroxylase-positive neurons in the iron-infused LC, indicating a reduction of neuron number by iron infusion. The involvement of apoptosis in iron-induced oxidative injuries was studied. An abrupt increase in cytosolic cytochrome c content was demonstrated in the infused LC 48 hours after iron infusion. TUNEL-positive cells, an indication of apoptosis, were detected in the iron-infused LC. In an attempt to prevent iron-induced neurotoxicity, vitamin D3, an active metabolite of vitamin D,

was systemically administered. Iron-induced increases in cytosolic cytochrome c and TUNEL-positive cells were reduced by this treatment. Furthermore, systemic administration of vitamin D3 attenuated iron-induced oxidative injuries in the infused LC. Our data suggest that local infusion of iron in LC induced oxidative stress and resulted in programmed cell death in the LC-hippocampal noradrenergic system. Furthermore, vitamin D3 may be neuroprotective and therapeutic in attenuating iron-induced neurotoxicity in CNS.

Chen KB, Lin AMY, Chiu TH. 2003. Oxidative injury to the locus coeruleus of rat brain: neuroprotection by melatonin. J Pineal Res 35(2):109-117.
Abstract: Neurodegeneration in the locus coeruleus (LC) has been documented in several central nervous system (CNS) neurodegenerative diseases. In the present study, iron-induced oxidative injury in the LC was investigated in chloral-hydrate anesthetized rats. Three days after bilateral infusion of iron in the LC, both vertical and horizontal locomotor activities were decreased. Seven days after unilateral infusion of iron, lipid peroxidation was elevated in the infused LC, and the norepinephrine content was depleted in the ipsilateral hippocampus of the brain. Furthermore, the immunohistochemical study demonstrated a reduction in tyrosine hydroxylase-positive neurons in the infused LC. The involvement of programmed cell death (apoptosis) in iron-induced oxidative injury in the LC was investigated. Forty-eight hours after iron infusion, cytosolic cytochrome c was elevated in the infused LC. Moreover, terminal deoxytransferase-mediated dUTP-nick end labeling (TUNEL)-positive cells, an indicative of apoptosis, were detected in the infused LC. In an attempt to prevent oxidative injury in the LC, melatonin was systemically administered. Intraperitoneal injection of melatonin attenuated iron-induced behavioral changes in locomotor activity as well as iron-induced increases in cytosolic cytochrome c and TUNEL-positive cells. Moreover, melatonin diminished iron-induced oxidative injury. At the same time, the level of glial derived neurotrophic factor (GDNF) was elevated in the LC of melatonin-treated rats. Our data suggests that oxidative stress because of iron results in apoptosis in the infused LC and causes degeneration of the coeruleohippocampal noradrenergic system in the rat brain. Furthermore, melatonin, among other mechanisms, may exert its neuroprotection via up-regulation of GDNF levels in CNS.

Chen CJ, Liao SL. 2003. Zinc toxicity on neonatal cortical neurons: involvement of glutathione chelation. J Neurochem 85(2):443-453.
Abstract: Several mechanisms have been implicated in pathological neuronal death including zinc neurotoxicity, calcium excitotoxicity and oxidative injury. Glutathione (GSH) serves to provide reducing equivalents for the maintenance of oxidant homeostasis, and also plays roles in intracellular and intercellular signaling in the brain. We investigated the role of GSH homeostasis in the neurotoxic action of zinc using both mixed cortical cultures containing neurons and glia, and cortical neurons prepared from 1-day-old rats. Zinc caused neuronal cell death in a concentration-dependent manner. In parallel, a high concentration of zinc depleted GSH, in a time-dependent manner, preceding the onset of neuronal damage. Depletion of GSH by diethylmaleate injured neurons and exacerbated zinc-induced death. In contrast, replenishment of GSH attenuated zinc neurotoxicity. The thiol-containing compounds N -acetylcysteine and GSH chemically chelated zinc leading to decreases in the influx of zinc, the fall in GSH level and neuronal death. Interestingly, the glycolytic substrate pyruvate, but not lactate, chelated zinc concentration dependently and prevented its toxicity. On the other hand, pyrrolidine dithiocarbamate, serving as a zinc chaperon, enhanced its entry and toxicity. The results suggest that zinc non-enzymatically depleted GSH, an intrinsic factor for neuron survival, leading to activation of the cellular death signal and eventually neuronal death.

Cantuti-Castelvetri I, Shukitt-Hale B, Joseph JA. 2003. Dopamine neurotoxicity: age-dependent behavioral and histological effects. Neurobiol Aging 24(5): 697-706.

Abstract: The oxidative stress (OS) theory has implicated the involvement of reactive oxygen species (ROS) in both aging and age-dependent neurodegenerative diseases. The dopaminergic system is particularly vulnerable to ROS, and dopamine (DA) itself can be an endogenous source of ROS. The present study evaluated the hypothesis that DA-induced toxicity is age-dependent, and tested the behavioral and histological correlates of DA neurotoxicity in aging. Young (6 months) and middle-aged (15 months) rats were chronically treated with DA in the substantia nigra (SN, 1 mumol/2 mul vehicle per side/day/5 days) and were subsequently examined for changes in motor function and histology. The neurotoxic effect of DA treatment was an age-dependent effect, as middle-aged animals that received DA infusions in the SN were more impaired than their age-matched controls, especially on tasks that involved greater sensory-motor coordination, whereas young animals that received DA behaved similarly to their age-matched controls. The behavioral effects noted were accompanied by a loss of the tyrosine hydroxylase phenotype in substantia nigra. However, selective neurodegeneration was not noted in the SN of the treated animals, nor was a selective iron deposition noted at the site of injection. These results suggest that a neurochemical deficit and not cell loss per se within the nigrostriatal system underlies the motor behavioral deficits observed in the middle-aged rats. (C) 2002 Elsevier Science Inc. All rights reserved.

Canfield RL, Kreher DA, Cornwell C, Henderson CR. 2003. Low-level lead exposure, executive functioning, and learning in early childhood. Child Neuropsychology 9(1):35-53.
Abstract: The current paper presents evidence relating low-level lead exposure to impaired executive functioning in young children. Using the Shape School task, we assessed focused attention, attention switching, working memory, and the ability to inhibit automatic responses in a cohort of 170 children. Participants performed the Shape School task at both 48 and 54 months of age; the mean blood lead level was 6.49 mug/dl at 48 months. After controlling for a wide range of sociodemographic, prenatal, and perinatal variables, blood lead level was negatively associated with children's focused attention while performing the tasks, efficiency at naming colors, and inhibition of automatic responding. In addition, children with higher blood lead levels completed fewer phases of the task and knew fewer color and shape names. There was no association between blood lead and performance on the most difficult tasks, those requiring attention switching or the combination of inhibition and switching. Children's IQ scores were strongly associated with blood lead and Shape School performance, and when entered as a covariate, only color knowledge and the number of tasks completed remained significant. Results provide only weak support for impaired executive functioning, but the deficits in color knowledge may indicate a primary sensory deficit or difficulty with forming conditional associations, both implicating disruptions in dopamine system function.

Burkhard PR, Delavelle J, Du Pasquier R, Spahr L. 2003. Chronic Parkinsonism associated with cirrhosis - A distinct subset of acquired hepatocerebral degeneration. Arch Neurol 60(4):521-528.
Abstract: Context: The clinical, neuroradiological, and biological characteristics of the so-called acquired hepatocerebral degeneration have not yet been fully determined and its frequency remains largely uncertain. Objectives: To prospectively study the prevalence of extrapyramidal symptoms in patients with moderate to. severe cirrhosis of various causes, to delineate the main neurological features of the condition, and to establish correlations with neuroradiological and biological findings. Patients and Methods: During a 1-year period, all consecutive patients with cirrhosis who were potential candidates for liver transplantation were screened for extrapyramidal features. When extrapyramidal features were present, further workup included a detailed neurological examination, magnetic resonance imaging of the brain, a comprehensive battery of neuropsychological tests, extensive blood tests, and, in some cases, cerebrospinal fluid analysis. Setting: A community-based hospital. Results:

From 51 patients screened, 11 (21.6%) exhibited moderate to severe parkinsonism sometimes associated with focal dystonia. Typical features included rapid progression over months, symmetric akinetic-rigid syndrome, postural but not resting tremor, and early postural and gait impairment. Neuropsychiatric manifestations were minimal. Some patients were responsive to levodopa therapy. In all patients, magnetic resonance imaging scans showed striking hyperintensities on T1-weighted images typically involving the substantia nigra and the globus pallidus bilaterally. Whole blood and cerebrospinal fluid manganese concentrations were severalfold above the reference range. Conclusions: Cirrhosis-related parkinsonism may represent a unique, consistent, and common subset of acquired hepatocerebral degeneration, whose features are permanent and entirely different from acute hepatic encephalopathy episodes. This form of parkinsonism can be clearly distinguished from other forms of parkinsonism of middle to advanced age, based on a suggestive association of clinical, neuroradiological, and biological abnormalities. Our findings support the concept of the toxic effects of manganese being the major determinant of basal ganglia dysfunction leading to the predominantly extrapyramidal central nervous system manifestations of cirrhosis observed in these patients.

Bostrom AC. 2003. Technologic advances in psychiatric nursing. Nurs Clin North Am 38(1):1-+.

Abstract: Historically, mental health care and psychiatric nursing used little technology. Psychiatric care, even up to the last 2 or 3 decades of the twentieth century, had little use for gadgets. Diagnosis was made on the basis of careful interview and deductive logic. No reliable laboratory tests confirmed these diagnoses consistently. Treatment used talking and medications and environmental management to help patients make improvements in their symptoms. When the author first started practicing in 1972, the technology used consisted of a small metal box with a few buttons and dials. It was located in the treatment room off the main hallway of the unit. There, rarely, the most intransigent of patients were taken. They were the patients who did not respond to the milieu, the talking, the group activities, or the medications. The team consisting of the physician, a nurse, an aide, and an anesthesiologist would administer a barbiturate and succinylcholine-and enough electric current to each temple to generate a grand mal seizure. A year later, the author was working in a different hospital, where electroconvulsive therapy was given to many patients. In a bright procedure room, depressed patients were scheduled every half hour or so for their procedure. With more neuromuscular blockade prescribed in the protocol at this facility, the seizures were less pronounced. From the procedure room, the patients were rolled into the recovery room, where two rows of patients recovered from their electroconvulsive therapy. Technology was not consistently embraced within mental health treatment. Psychiatric care in the early twentieth century used mostly mechanical technology if any was used at all. Treatment before the discovery of effective psychoactive medications included restraint devices (eg, straitjackets, sheet wraps that were often wet, and leather restraints), baths, seizures produced by insulin or electric shock treatment, and barbiturate or soporific medications (eg, chloral hydrate and paraldehyde). The causes of mental illness were explored through history taking or psychoanalytic methods. At most, the technology of gross anatomic dissections of brains of mentally ill patients revealed a few things: The ventricles of patients with schizophrenia are larger than those of nonschizophrenic patients, and the brain itself is atrophied. Most brains came from individuals who had been ill for many years or who were elderly. It was difficult to sort to what degree these brain changes were explained by the disease alone. Medications that improved symptoms (more than just putting patients to sleep) were discovered in the mid-twentieth century. Scientists then learned a great deal about the brain from animals [1], in which specific areas of the brain were destroyed, psychoactive medications were given, and behavioral manifestations that were eliminated or enhanced were measured. As a result of this information, combined with the pharmacologic and neurochemical

knowledge at the time, scientists learned about the location and types of neurotransmission in the brain. Disorders in humans caused by known brain deficiencies, such as Parkinson's disease, suggested hypotheses about the mechanisms of action of drugs that created behavioral syndromes similar to the deficiency syndromes. This research culminated at the end of the twentieth century with the "decade of the brain," during which there was exponential growth in the understanding of the brain and technologies for understanding the brain. At the same time, a major shift in the attitude toward psychiatric care, patients, and practitioners was occurring. Psychiatric symptoms were less stigmatizing to the patient. Families could point to the biology of the illness rather than to the "schizophrenogenic mother" or family. Psychiatrists could rejoin, with legitimacy, their colleagues in other specialties who primarily treated their patients with medications or surgery. Technology changed the lives of everyone involved in the mental health system directly and indirectly. This article describes some of the technologies available for research, assessment, and treatment, including a variety of imaging techniques, genetic explorations, medications, and alternative treatments. What these technologies mean for nurses working with the mentally ill is discussed briefly.

Bostanci MO, Bagirici F, Korkmaz A. 2003. The neurotoxic effect of iron on pyramidal cell number in rat hippocampus: A stereological study. Neuroscience Research Communications 32(3):151-159.
Abstract: There have been numerous studies of neuronal hyperactivity and oxidative stress induced by iron. Moreover, it has been found that iron levels in the brain are markedly increased in some neurodegenerative diseases such as Alzheimer's disease (AD) and Parkinson's disease (PD). We attempted to identify the toxic effect of iron on the hippocampus, a region of the brain known to be involved in learning and memory processes and which lends itself to stereological analysis. Rats were given iron chloride (FeCl(3)5H(2)O, 200 mM in 2.5 mul) intracortically and were killed 10 days later. The total number of neurons in the hippocampus of control and iron-treated rats was counted with an optical fractionator. Iron administration caused a significant decrease in the total number of pyramidal neurons in the left (31.6% decrease) and right (32.2% decrease) hippocampi ($p<0.001$). The findings suggest that excess iron contributes to pyramidal cell death in the rat hippocampus.

Bocca B, Alimonti A, Forte G, Petrucci F, Pirola C, Senofonte O, Violante N. 2003. High-throughput microwave-digestion procedures to monitor neurotoxic elements in body fluids by means of inductively coupled plasma mass spectrometry. Analytical and Bioanalytical Chemistry 377(1):65-70.
Abstract: Microwave (MW) digestion procedures with high sample throughput (simultaneous digestion of 36 or 80 samples) and procedural simplicity (disposable plastic tubes, or re-usable liners with screw-cap) were investigated for their efficiency in routine analyses of biological samples. Different digestion vessel materials were tested for metal leaching/adsorption and thermal resistance: quartz, glass, polyethylene (PE) and polystyrene (PS). For the instrumental quantification of Al, Bi, Cd, Co, Cr, Hg, Mn, Mo, Ni, Pb, Sb, and Tl at ultra-trace levels in urine, serum, and whole blood, sector field inductively coupled plasma mass spectrometry (SF-ICP-MS) was used. The different pretreatment conditions and vessels were evaluated in terms of contamination risk, effective power of detection, accuracy, and precision. Results of analyses of serum, urine and whole blood certified reference materials (CRMs) were fully satisfactory for almost all the analytes. In the case of Hg, Mo, and Tl in serum digested in plastic containers the results were just below the lower limit of uncertainty of the certified range. On the basis of the present data the following MW procedures can be suggested: 1.for urine, digestion with nitric acid at atmospheric pressure in plastic vials; 2.for serum, digestion with nitric acid at atmospheric pressure in glass vessels; and 3.for whole blood, digestion under pressure in quartz tubes. Because of the levels of the procedural blanks, Bi was not measurable at the concentrations expected in human fluids, and Al was accurately detectable in whole blood

only.

Behnke S, Berg D, Becker G. 2003. Does ultrasound disclose a vulnerability factor for Parkinson's disease? J Neurol 250:24-27.
Abstract: Transcranial ultrasound is a new tool allowing the detection of abnormalities in the echomorphology of the substantia nigra (SN) in patients with Parkinson's disease (PD). Several lines of evidence suggest that the changes in the echo-pattern represent a risk factor as: i) the majority of PD patients exhibit this echo-feature, ii) the presence of such changes in healthy controls is related to a reduced F-18-Dopa-uptake and clinical signs of nigrostriatal dysfunction. The reason for the change of echogenicity is suggested to be an increased iron content in the substantia nigra causing oxidative stress and neuronal cell damage. This hypothesis of changes in SN echomorphology reflecting a risk factor of PD has to be proved in longitudinal studies.

Beal MF. 2003. Bioenergetic approaches for neuroprotection in Parkinson's disease. Ann Neurol 53:S39-S47; discussion S47-S48.
Abstract: There is considerable evidence suggesting that mitochondrial dysfunction and oxidative damage may play a role in the pathogenesis of Parkinson's disease (PD). This possibility has been strengthened by recent studies in animal models, which have shown that a selective inhibitor of complex I of the electron transport gene can produce an animal model that closely mimics both the biochemical and histopathological findings of PD. Several agents are available that can modulate cellular energy metabolism and that may exert antioxidative effects. There is substantial evidence that mitochondria are a major source of free radicals within the cell. These appear to be produced at both the iron-sulfur dusters of complex I as well as the ubiquinone site. Agents that have shown to be beneficial in animal models of PD include creatine, coenzyme Q(10), Ginkgo biloba, nicotinamide, and acetyl-L-carnitine. Creatine has been shown to be effective in several animal models of neurodegenerative diseases and currently is being evaluated in early stage trials in PD. Similarly, coenzyme Q(10) is also effective in animal models and has shown promising effects both in clinical trials of PD as well as in clinical trials in Huntington's disease and Friedreich's ataxia. Many other agents show good human tolerability. These agents therefore are promising candidates for further study as neuroprotective agents in PD.

Barthel H, Hermann W, Kluge R, Hesse S, Collingridge DR, Wagner A, Sabri O. 2003. Concordant pre- and postsynaptic deficits of dopaminergic neurotransmission in neurologic Wilson disease. American Journal of Neuroradiology 24(2):234-238.
Abstract: BACKGROUND AND PURPOSE: Although previous brain imaging studies of Wilson disease (WD) focused on the dopaminergic system, correlational data on the integrity of the pre- and postsynaptic compartments are lacking. The present study was initiated to intraindividually determine the integrity of these compartments in patients with WD. METHODS: A total of 46 patients with WD and 10 matched control subjects underwent [I-123] 2beta-carbomethoxy-3beta(4[I-123] iodophenyl)tropane ([I-123]beta-CIT) and [I-123]iodobenzamide ([I-123] IBZM) single photon emission CT (SPECT). For both radiotracers, specific striatal binding ratios (with the cerebellum as the reference region) were calculated after a standardized region-of-interest technique was applied. In addition, the severity of putative neurologic symptoms was evaluated by using a linear scoring system. RESULTS: In patients without neurologic symptoms, striatal binding ratios of both radiotracers did not differ from those of the control group (13.8 +/- 3.1 vs 12.0 +/- 3.4 and 2.00 +/- 0.19 vs 1.90 +/- 0.27; n.s.). In symptomatic patients, however, striatal binding ratios for both [I-123]beta-CIT and [I-123]IBZM were significantly reduced (9.1 +/- 2.3 and 1.64 +/- 0.18; P <.001). In all patients with WD, the [I-123]β-CIT and [I-123] IBZM binding ratios were significantly correlated (r = 0.65, P <.001), as were SPECT parameters and the severity of the neurologic symptoms (r = -0.60 and -0.62; P <.001). CONCLUSION: These findings of a concordant bicompartmental

dopaminergic deficit in neurologic WD provide in vivo evidence for assigning WD to the group of secondary Parkinsonian syndromes. These results could be relevant in therapeutic decision making in patients with this copper deposition disorder.

Barreto WJ, Barreto SRG, Kawano Y, Deoliveira LFC, Di Mauro E, Paschoal FMM. 2003. Preparation and spectroscopic characterization of two manganese(II) semiquinone complexes. Monatshefte Fur Chemie 134(12):1545-1554.
Abstract: The complexes [CTA][Mn(II)(SQ)(3)] were isolated in the solid state and purified. SQ is the o-semiquinone of L-dopa or dopamine and CTA is the cetyltrimethylammonium cation. These complexes were characterized by Raman, infrared, EPR and thermogravimetry (TG) techniques. The EPR spectra of the solids presented an intense signal characteristic of the o-semiquinone radical anion with g=2.0062 and g=2.0063 for L-dopa and dopamine, respectively. Six characteristic lines around the organic radical signal confirm the presence of the Mn2+ ion. The most intense Raman bands were observed at (ν) over bar for dopamine and at 1356 cm(-1) for L-dopa and assigned to a C-O stretching with major C-1-C-2 character. The absence of an intense Raman band at ca. (ν) over bar = 1480 cm(-1), characterizes the ligands as an o-semiquinone radical anion. Broad bands in the (ν) over bar = 400-750 cm(-1) region can be assigned to deformations associated with the five-member ring chelate including the manganese ion, the oxygens, and the C-1-C-2 bonds. The more intense IR bands for the dopamine and the L-dopa-derived ligands at (ν) over bar = 1233 and 1229 cm(-1) are assigned to (ν) over bar CO. Mass loss mechanisms for the two complexes, based on the TG results, were proposed and confirm the formula proposed.

Baek SY, Lee MJ, Jung HS, Kim HJ, Lee CR, Yoo C, Lee JH, Lee H, Yoon CS, Kim YH, Park J, Kim JW, Joen BS, Kim Y. 2003. Effect of manganese exposure on MPTP neurotoxicities. Neurotoxicology 24(4-5):657-665.
Abstract: We used a 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP)-treated mice model to evaluate whether manganese (Mn) exposure can affect MPTP-induced neurotoxicity. We randomly assigned adult male C57BL/6 mice (n = 5-7 per group) the following treatments: SO, Mn(-) MPTP(-); MO, Mn(+) MPTP(-); SM, Mn(-) MPTP(+); MM, Mn(+) MPTP(+). Mn (MnCl(2)4H(2)O) was administered intraperitoneally at a dose of 2 mg/kg daily for 3 weeks. MPTP was then administered intraperitoneally at a dose of 30 mg/kg daily for 5 days in the SM and MM groups. Seven days after the last MPTP injection, the animals were sacrificed. Blood Mn levels were elevated in the Mn-exposed groups. Striatal Mn levels were not influenced by Mn treatment alone, however they were decreased following MPTR Tyrosine hydroxylase (TH)-immunoreactive (ir) neurons in the substantia nigra pars compacta (SNpc) were decreased significantly in the MPTP-exposed groups. Densities of TH- and dopamine transporter (DAT)-ir axon terminals in the caudate-putamen (CPU) were also decreased in the MPTP-treated groups. Furthermore, glial fibrillary acidic protein (GFAP)-ir astrocytes increased in the CPU with MPTP treatment. However no effects were observed with Mn exposure. Concentrations of dopamine (DA), 3,4-dihydrophenyl acetic acid (DOPAC) and homovanillic acid (HVA) in the corpus striatum were also decreased significantly with MPTP treatment alone, but Mn had no effect. Thus, decreased dopaminergic activities with MPTP led to decreased DA and its metabolites. Significant hypertrophies of GFAP-ir astrocytes in the globus pallidus (GP) were observed in Mn-exposed groups, especially in the MM group. MPTP targeted dopaminergic systems whereas Mn neurotoxicities occurred in the GP In conclusion, our data suggest that Mn does not potentiate the neurotoxicity of MPTP. (C) 2003 Elsevier Science Inc. All rights reserved.

Atorino L, Silvestri L, Koppen M, Cassina L, Ballabio A, Marconi R, Langer T, Casari G. 2003. Loss of m-AAA protease in mitochondria causes complex I deficiency and increased sensitivity to oxidative stress in hereditary spastic paraplegia. J Cell Biol 163(4):777-787.
Abstract: Mutations in paraplegin, a putative mitochondrial

metallopeptidase of the AAA family, cause an autosomal recessive form of hereditary spastic paraplegia (HSP). Here, we analyze the function of paraplegin at the cellular level and characterize the phenotypic defects of HSP patients' cells lacking this protein. We demonstrate that paraplegin coassembles with a homologous protein, AFG3L2, in the mitochondrial inner membrane. These two proteins form a high molecular mass complex, which we show to be aberrant in HSP fibroblasts. The loss of this complex causes a reduced complex I activity in mitochondria and an increased sensitivity to oxidant stress, which can both be rescued by exogenous expression of wild-type paraplegin. Furthermore, complementation studies in yeast demonstrate functional conservation of the human para-plegin-AFG3L2 complex with the yeast m-AAA protease and assign proteolytic activity to this structure. These results shed new light on the molecular pathogenesis of HSP and functionally link AFG3L2 to this neurodegenerative disease.

Archer T, Schroder N, Fredriksson A. 2003. Neurobehavioural deficits following postnatal iron overload: II instrumental learning performance. Neurotoxicity Research 5(1-2):77-94.
Abstract: Three experiments reporting instrumental learning deficits following postnatal iron overload in rodents are described. In Experiment 1, NMRI mice were given different doses of iron succinate (0.0, 3.7 or 37.0 mg Fe2+/kg b.w., p.o.) on different postnatal days (PD). In the PD 10-12 group, there were marked disruptions of radial arm maze learning performance in the 37.0 mg Fe2+/kg dose group, and to a lesser extent in the 3.7 mg Fe2+/kg dose group. In Experiment II, newborn NMRI mice were administered Fe2+ (7.5 mg/kg, b.w.) at either PD 3-5, PD 10-12, or PD 19-21, or vehicle (saline). Marked deficits in radial arm maze performance were obtained in the mice administered iron during PD 1012, and to a much lesser extent PD 3-5. In Experiments III and IV, rats were administered two different dose regimes: (i) Experiment III: 2.5, 7.5, 15.0 or 30 mg Fe2+/kg b.w., p.o., at PD 10-12, or (ii) Experiment IV 2.5, 7.5 or 22.5 mg Fe2+/kg b.w., p.o., at PD 10-12, followed 3 months later by behavioural testing. All four dose groups in Experiment III demonstrated marked deficits in radial arm maze learning and retention performance but only the 30 mg/kg group showed hypoactivity. In Experiment IV, deficits in inhibitory conditioning were obtained in the 7.5 and 15.0, but not the 2.5, mg/kg dose groups. The analysis of enzymes involved in oxidative stress indicated that: (1) Formation of thiobarbiturate acid reactive species (TBARS) concentration was elevated in the substantia nigra by both the 7.5 and 15.0 mg Fe2+/kg doses whereas in the striatum there was a decrease. (2) Superoxide dismutase activity was decreased in a dose-related fashion in the substantia nigra but was elevated in the cerebellum. Iron-overload during the immediate postnatal period incorporating critical synaptogenesis seems detrimental for several aspects of functional and neurobiological development.

[Anon]. 2003. Iron intake possibly linked to increased risk of Parkinson's disease. Cns Spectrums 8(7):482.

[Anon]. 2003. Iron: a causal factor in Parkinson's disease? Neuroscientist 9(4): 236.

Andersen JK. 2003. Paraquat and iron exposure as possible synergistic environmental risk factors in Parkinson's disease. Neurotoxicity Research 5 (5):307-313.

Adams LA, Aggarwal VK, Bonnert RV, Bressel B, Cox RJ, Shepherd J, De Vicente J, Walter M, Whittingham WG, Winn CL. 2003. Diastereoselective synthesis of cyclopropane amino acids using diazo compounds generated in situ. J Org Chem 68(24):9433-9440.
Abstract: A simple and high-yielding method for the preparation of cyclopropane amino acids is described. The novel method involves the one-pot cyclopropanation of readily available dehydroamino acids using aryl and unsaturated diazo compounds generated in situ from the

corresponding tosylhydrazone salts. It was found that thermal 1,3-dipolar cycloaddition followed by nitrogen extrusion gave the cyclopropane amino acid derivatives with good E selectivity, while reactions in the presence of meso-tetraphenylporphyrin iron chloride gave predominantly the corresponding Z isomers. The synthetic utility of this process was demonstrated in the synthesis of (+/-)-(Z)-2,3-methanophenylalanine [(+/-)-(Z)-1], the anti-Parkinson (+/-)-(E)-2,3-methano-m-tyrosine [(+/-)-(E)-2], and the natural product (+/-)-coronamic acid [(+/-)-3].

Abduljalil AM, Schmalbrock P, Novak V, Chakeres DW. 2003. Enhanced gray and white matter contrast of phase susceptibility-weighted images in ultra-high-field magnetic resonance imaging. J Magn Reson Imaging 18(3):284-290.
Abstract: Purpose: To evaluate if magnetic susceptibility sensitive phase postprocessed images can be used to enhance the inherent brain/gray white matter contrast in gradient echo (GE) images at 8-Tesla (T) magnetic resonance (MR). Materials and Methods: Phase and magnitude images of high-resolution GE MR 8-T images were created. Comparisons were made between the magnitude; the product of the magnitude and phase, and pure phase images. Results: The pure phase images significantly improved the contrast between the gray and white matter structures. In general, the higher the iron content or subvoxel field inhomogeneities, the higher was the contrast, and the greater were the resultant phase shifts. The phase images best demonstrated anatomy that was not apparent on the standard magnitude images. Conclusion: Phase imaging can significantly improve the demonstration of the internal anatomical brain structures over standard magnitude GE imaging techniques at high field.

Herrero Hernandez E, Valentini MC, Discalzi G. 2002 Dec. T1-weighted hyperintensity in basal ganglia at brain magnetic resonance imaging: are different pathologies sharing a common mechanism? Neurotoxicology 23 (6):669-74.
Abstract: Basal ganglia bilateral symmetric hyperintensity in T1-weighted sequences at magnetic resonance imaging (MRI) is recognized to be due to the presence of manganese deposits. This abnormal finding has been reported in occupational exposures, liver cirrhosis and total parenteral nutrition with unbalanced solutions. However, the same imaging is often observed "by chance" in brain MRIs of patients not belonging to these groups. In order to better understand which are the clinical conditions coexisting with such findings, we decided to study systematically patients which showed this kind of imaging, focusing on their manganese and iron status, as it is known that these two metals have similar properties and that iron-deficiency can competitively increase manganese absorption. The 20 patients studied underwent clinical evaluation and the following laboratory tests: whole blood iron and manganese, hemoglobin, plasma iron, transferrin and ferritin. The neuroradiologic evaluation was integrated by pallidal index calculation, in order to provide a semi-quantitative esteem of the hyperintensity. The patients could be classified into four subgroups: Parkinsonism, anemia, cirrhosis, central nervous system tumors. In 18 out of 20 cases, we found abnormalities in iron and/or manganese-related values. Particularly, iron-deficiency seems to be frequent among patients showing brain MRI abnormalities compatible with manganese deposits in basal ganglia. This observation suggests that iron-deficiency could be an important risk factor for manganese-induced neurotoxicity and should, therefore, be accurately considered and treated.

Youdim MB, Grunblatt E, Levites Y, Maor G, Mandel S. 2002 Nov. Early and late molecular events in neurodegeneration and neuroprotection in Parkinson's disease MPTP model as assessed by cDNA microarray; the role of iron. Neurotox Res 4(7-8):679-689.
Abstract: Possible cell death mechanisms for pars compacta nigro-striatal dopamine neurons in Parkinson's disease include oxidative stress, inflammatory processes, nitric oxide iron accumulation, glutamate toxicity and diminished neurotrophic factor responses. There is a notion that Parkinson's disease is not a single disorder but a syndrome that can be initiated by several factors. Because of limitations of biochemical methods

in the global analysis of neuronal death, a full picture of events has not been established. However, recently developed cDNA microarray or microchips, in which the global expression of thousands of genes can be assessed simultaneously, is changing the prospect for understanding the disease process, its progression, response to drugs, etc. The neurotoxin N-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) is considered the most valid model of Parkinson's disease. We employed the technique of cDNA microarray gene expression to determine the mechanism of action of MPTP in mouse substantia nigra. Also, we studied neuroprotective processes induced by several compounds, including R-apomorphine and the green tea polyphenol epigallo-catechin-3-gallate (EGCG). This was done in two ways: (1) the time-dependent acute effect of MPTP, for determining which of the initial genes might lead to dopamine neuron death and (2) gene expression at the time of MPTP-induced dopamine neuron death. We observed that early (acute MPTP) gene expression differs from effects seen at the time of death (chronic MPTP), and that early gene changes are crucial for setting into action genes that eventually cause dopamine neuron death. Furthermore, this process is a cascade of "domino" effects, some of which were previously established by biochemical means. However, our findings show an additional large number of events previously unknown. The neuroprotective drugs reversed some but not all of the gene expression, suggesting involvement of these genes in the neurodegenerative process. Because of the profound complexity of "domino" effect it is now reasonable to understand why a single neuroprotective drug has not shown clinical neuroprotective efficacy. Future multi neuroprotective drugs may be necessary for treatment of not only Parkinson's disease, but other neurodegenerative diseases (e.g. Alzheimer's disease) and detrimental states (e.g. ischaemia).

Weiss B, Clarkson TW, Simon W. 2002 Oct. Silent latency periods in methylmercury poisoning and in neurodegenerative disease. Environ Health Perspect 110 Suppl 5:851-4.
Abstract: This article discusses three examples of delay (latency) in the appearance of signs and symptoms of poisoning after exposure to methylmercury. First, a case is presented of a 150-day delay period before the clinical manifestations of brain damage after a single brief (<1 day) exposure to dimethylmercury. The second example is taken from the Iraq outbreak of methylmercury poisoning in which the victims consumed contaminated bread for several weeks without any ill effects. Indeed, signs of poisoning did not appear until weeks or months after exposure stopped. The last example is drawn from observations on nonhuman primates and from the sequelae of the Minamata, Japan, outbreak in which low chronic doses of methylmercury may not have produced observable behavioral effects for periods of time measured in years. The mechanisms of these latency periods are discussed for both acute and chronic exposures. Parallels are drawn with other diseases that affect the central nervous system, such as Parkinson disease and post-polio syndrome, that also reflect the delayed appearance of central nervous system damage.

Herbst T, Anis-Anwar Y, Giacco S, White WB. 2002 Oct. Evaluation of the overall system precision of the Welch-Allyn transtelephonic home blood pressure monitor in adults with Parkinson's disease. Blood Press Monit 7(5):285-8.
Abstract: BACKGROUND: Non-invasive blood pressure (BP) devices should be independently evaluated before being used in special populations. The objective of this study was to assess the accuracy of the Welch-Allyn transtelephonic home blood pressure monitor in adults with Parkinson's disease to evaluate the device for use in a large clinical trial involving the safety and efficacy of a monoamine oxidase inhibitor. METHODS: BP measurements taken with the device were compared with the results obtained by two experienced observers using a mercury sphygmomanometer in patients with Parkinson's disease. The limits of agreement were then calculated for the device and compared with the results of the two observers. RESULTS: The agreement parameters between the two observers were -0.5 +/- 2.6 mmHg for systolic BP and 0.1 +/- 2.2 mmHg for diastolic BP. The agreement between the Welch-Allyn

transtelephonic device and the observers was -2.6 +/- 4.5 mmHg and -1.9 +/- 3.2 mmHg for systolic and diastolic BP respectively. Nearly 90% of the readings were within 10 mmHg of the observers for both systolic and diastolic BP. Mild tremor had a moderate effect on the validity of the device. CONCLUSIONS: The Welch-Allyn transtelephonic device demonstrated acceptable precision in this cohort of patients with Parkinson's disease and is considered valid for use in a clinical trial involving these patients.

Becker G, Muller A, Braune S, Buttner T, Benecke R, Greulich W, Klein W, Mark G, Rieke J, Thumler R. 2002 Oct. Early diagnosis of Parkinson's disease. J Neurol 249 Suppl 3:III/40-8.
Abstract: In idiopathic Parkinson's disease (IPD) approximately 60 % of the nigrostriatal neurons of the substantia nigra (SN) are degenerated before neurologists can establish the diagnosis according to the widely accepted clinical diagnostic criteria. It is conceivable that neuroprotective therapy starting at such an 'advanced stage' of the disease will fail to stop the degenerative process. Therefore, the identification of patients at risk and at earlier stages of the disease appears to be essential for any successful neuroprotection. The discovery of several genetic mutations associated with IPD raises the possibility that these, or other biomarkers, of the disease may help to identify persons at risk of IPD. Transcranial ultrasound have shown susceptibility factors for IPD related to an increased iron load of the substantia nigra. In the early clinical phase, a number of motor and particularly non-motor signs emerge, which can be identified by the patients and physicians years before the diagnosis is made, notably olfactory dysfunction, depression, or 'soft' motor signs such as changes in handwriting, speech or reduced ambulatory arm motion. These signs of the early, prediagnostic phase of IPD can be detected by inexpensive and easy-to-administer tests. As one single instrument will not be sensitive enough, a battery of tests has to be composed measuring independent parameters of the incipient disease. Subjects with abnormal findings in this test battery should than be submitted to nuclear medicine examinations to quantify the extent of dopaminergic injury and to reach the goal of a reliable, early diagnosis.

Noor R, Mittal S, Iqbal J. 2002 Sep. Superoxide dismutase--applications and relevance to human diseases. Med Sci Monit 8(9):RA210-5.
Abstract: Reactive oxygen species, such as superoxide radicals, are thought to underlie the pathogenesis of various diseases. Almost 3 to 10% of the oxygen utilized by tissues is converted to its reactive intermediates, which impair the functioning of cells and tissues. Superoxide dismutase (SOD) catalyzes the conversion of single electron reduced species of molecular oxygen to hydrogen peroxide and oxygen. There are several classes of SOD that differ in their metal binding ability, distribution in different cell compartments, and sensitivity to various reagents. Among these, Cu, Zn superoxide dismutase (SOD1) is widely distributed and comprises 90% of the total SOD. This ubiquitous enzyme, which requires Cu and Zn for its activity, has great physiological significance and therapeutic potential. The present review describes the role of SODs, especially Cu, Zn SOD, in several diseases, such as familial amyotrophic lateral sclerosis (FALS), Parkinson's disease, Alzheimer's disease, dengue fever, cancer, Down's syndrome, cataract, and several neurological disorders. Mutations in the SOD1 gene cause a familial form of amyotrophic lateral sclerosis. The mechanism by which mutant SOD1 causes the degeneration of motor neurons is not well understood. Transgenic mice expressing multiple copies of FALS-mutant SOD1s develop an ALS-like motor neuron disease. Vacuolar degeneration of mitochondria has been identified as the main pathological feature associated with motor neuron death and paralysis in several lines of FALS-SOD1 mice. Various observations and conclusions linking mutant SOD1 and FALS are discussed in this review in detail.

Normandin L, Panisset M, Zayed J. 2002 Jul-Sep. Manganese neurotoxicity: behavioral, pathological, and biochemical effects following various routes

of exposure. Rev Environ Health 17(3):189-217.
Abstract: The human central nervous system is an important target for manganese intoxication, which causes neurological symptoms similar to those of Parkinson's disease. With the increasing use of methylcyclopentadienyl manganese tricarbonyl (MMT) as an octane-improving additive to unleaded gasoline, the prospect of worldwide manganese exposure is once again attracting attention as increases in environmental manganese concentrations have been recorded relative to traffic density. One crucial question is whether a small increase of manganese contamination resulting from the widespread use of MMT could have neurotoxic effects. In this review we concentrate on central nervous system abnormalities and neurobehavioral disturbances. Most experimental animal studies on manganese neurotoxicity have been conducted in nonhuman primates and rodents. Most studies performed in rodents used oral manganese administration and did not assess bioaccumulation or central nervous system changes. The major effect found was transient modification of spontaneous motor activity. Very few inhalation toxicological studies were carried out. As manganese intoxication in humans usually occurs via inhalation, more studies are required using the respiratory route of administration. Given the proven neurotoxic effects of manganese and the prospect of worldwide MMT usage, this metal should be considered a new environmental pollutant having potentially widespread public health consequences.

Kurisaki H, Yomono H, Murayama S, Hebisawa A . 2002 Apr. [Multiple system atrophy with a-/hypo-ceruloplasminemia: distribution of iron in brains of 2 autopsy cases]. Rinsho Shinkeigaku 42(4):293-8.
Abstract: OBJECTIVE: We presented first two cases of multiple system atrophy (MSA) with a-/hypo-ceruloplasminemia (hypo-Cp). To know whether hypo-Cp was a cause of MSA, we investigated distribution of iron in brains. METHODS: Investigating history, neurological sings and symptoms, neuroimagings, and neuropathological findings of the 2 cases, we demonstrate that these 2 cases were typical MSA. Serum ceruloplasmin (Cp) values of two cases were investigated, as well as those of 14 MSA patients diagnosed after the 2 cases. In the 2 cases, we compared distribution of lesions and distribution of iron depositions revealed by Berlin blue stain (iron stain). Further, we compared depositions of iron in substantia nigra, putamen, and dentate nucleus of the 2 cases with those of 4 control MSA, 2 Parkinson's disease (PD), 2 amyotrophic lateral sclerosis (ALS), and 3 controls. RESULTS: Case 1 was 68-year-old man who developed gait disturbance, and had anti-Parkinson disease drugs after diagnosis of PD. Parkinsonism was progressed, and became bed-ridden after 6 years when he died. Neuropathological finding was typical MSA from distribution of lesions, as well as existence of GCIs and NCIs. Case 2 was 61-year-old man who developed parkinsonism. After 9 years, he had tracheostomy, and after 11 years died of renal failure. Neuropathological finding was typical MSA. With an investigation of serum Cp values of clinically diagnosed 14 MSA patients, we found 2 other cases of MSA with hypo-Cp. Iron stain of the 2 brains revealed that iron depositions were found in substantia nigra and putamen, but were not found neither in pontine base, cerebellum, nor inferior olive. Iron depositions were also seen in substantia nigra and putamen of control MSA cases as same degree as MSA with hypo-Cp, but iron depositions were fewer in PD, ALS and controls. CONCLUSION: Clinico-pathological findings of the the 2 cases were those of typical MSA, but were not same as those of previously reported hypo-Cp. Previous reports suggested iron depositions as a cause of brain lesions, but, we concluded that, in the 2 cases, iron depositions were not a direct cause of MSA lesions. However, high incidence of association of hypo-Cp and MSA shown in our study suggests a relation between hypo-Cp and MSA.

Gatto EM, Riobo N, Carreras MC, Poderoso JJ, Micheli FE. 2002 Mar. Neuroprotection in Parkinson's disease; a commentary. Neurotox Res 4(2): 141-5.
Abstract: Parkinson's disease (PD) is a worldwide neurodegenerative

disorder. Although the etiology has been linked to genetic and environmental factors, curative treatment remains a challenge. Several hypotheses support different pathophysiological mechanisms related to oxidative stress, glutamate-mediated neurotoxicity, mitochondrial energetic impairment and nitric oxide (NO) over-production. Moreover, apoptotic mechanisms have been identified in PD. In this way, classical drugs such as amantadine, selegiline and dopamine agonists show only a modest neuroprotective effect. New strategies with enormous potential are now under development. These include neuroprotectants and agents that might rescue dopaminergic neurons. Glutamate receptor antagonists, neurotrophins, neuroimmunophilins, adenosine A2A receptor antagonists, iron-chelators and NO modulators, as well as caspase inhibitors have evident neuroprotective properties in experimental PD models.

Le Couteur DG, Muller M, Yang MC, Mellick GD, McLean AJ. 2002 Jan-Mar. Age-environment and gene-environment interactions in the pathogenesis of Parkinson's disease. Rev Environ Health 17(1):51-64.
Abstract: Parkinson's disease (PD) is a common neurodegenerative disease characterized by dopaminergic cell death and deposition of Lewy bodies within the substantia nigra of the midbrain. Although the major risk factors for PD are aging and environmental factors, there is an important genetic component. An age-related change in xenobiotic metabolism alters the metabolism of and net exposure to, environmental neurotoxins. Genetic variability in xenobiotic metabolism may similarly increase the susceptibility to PD by altering the metabolism of neurotoxins. Genetic studies of rare familial cases of PD indicate a central mechanistic role for the aggregation of alpha-synuclein, a protein found in Lewy bodies. Environmental factors like pesticides and heavy metals can also influence alpha-synuclein aggregation. Common final pathways for aging, environmental, and genetic mechanisms can thus exist, involving both direct neurotoxicity and alpha-synuclein aggregation.

Castaneda MA, Ubilluz R, Avalos C, Escalante D, Nicoll J. 2002 Jan-Mar. [Wilson'S disease: dominant neuropsychiatric form. Case presentation and its physiopathologic interpretation based upon magnetic resonance of the encephalon]. Rev Gastroenterol Peru 22(1):74-80.
Abstract: This is a presentation of a clinical case of Wilson s disease. The patient is a 26 year old woman who began to evidence psychological symptoms, which were later accompanied by neurological manifestations such as asymmetrical hand tremor, parkinsonism, dystonia and later on, dysphagia and mutism. The ophthalmological examination found a Kayser Fleischer ring in Descemet s membrane. There was disturbance of copper metabolism documented with reduction of serum ceruloplasmin and increase of the urinary excretion of copper. Cirrhosis was demonstrated through laparoscopy and liver biopsy. The brain magnetic resonance showed frontotemporal atrophy and a degenerative process at the basal ganglia, cerebellum and brain stem. This information could suggest probable neuropsychiatric physiopathology. The stenosis and intense cervical dysphagia, associated with the crycopharyngeal membrane, has not been mentioned previously.

Peng XR, Jia Z, Zhang Y, Ware J, Trimble WS. 2002 Jan. The septin CDCrel-1 is dispensable for normal development and neurotransmitter release. Mol Cell Biol 22(1):378-87.
Abstract: Septins are GTPases required for the completion of cytokinesis in a variety of organisms, yet their role in this process is not known. Septins may have additional functions since the mammalian septin CDCrel-1 is predominantly expressed in the nervous system, a largely postmitotic tissue. While relatively little is known about the function of this protein, we have previously shown that it is involved in regulated secretion. In addition, the gene encoding this protein maps to a locus often deleted in velo-cardiofacial and DiGeorge syndromes, and CDCrel-1 has recently been shown to be a direct target of the E3 ubiquitin ligase activity of Parkin, a causative agent in autosomal recessive forms of Parkinson's disease. Here we show that CDCrel-1 expression rises at the time of

synaptic maturation and that CDCrel-1 is present in a complex that includes the septins Nedd5 and CDC10. To investigate its function in the nervous system, we generated homozygotic CDCrel-1 null mice and showed that these mice appear normal with respect to synaptic properties and hippocampal neuron growth in vitro. Moreover, we found that while the expression of a number of synaptic proteins is not affected in the CDCrel-1 mutant mice, the expression of other septins is altered. Together, these data suggest that CDCrel-1 is not essential for neuronal development or function, and that changes in expression of other septins may account for its functional redundancy.

Zheng YX, Chan P, Pan ZF, Shi NN, Wang ZX, Pan J, Liang HM, Niu Y, Zhou XR, He FS. 2002. Polymorphism of metabolic genes and susceptibility to occupational chronic manganism. Biomarkers 7(4):337-346.
Abstract: In this study we investigated genetic polymorphisms of five metabolizing genes and their association with occupational chronic manganism. We recruited 49 patients with chronic manganism and 50 unrelated healthy control subjects who were welders and ferromanganese smelters and occupationally exposed to manganese dust and fume in the same workshops from three metallurgical industries. The controls were matched to the cases by sex, age, cigarette and alcohol intake, as well as the manganese exposure duration. Polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP) was used to genotype the cytochrome P450 2D6L gene (CYP2D6L) and the NAD(P) H: quinone oxidoreductase gene (NQO1). Allele-specific PCR was used to detect the cytochrome P450 1A1 gene (CYP1A1), and the glutathione-S-transferase mu and theta genes (GSTM and GSTT). The frequency of polymorphic alleles, a mutation of CYP2D6L, was significantly lower in patients with chronic manganism (16.3%) than in controls (29.0%). Individuals with the homozygote polymorphism (L/L) of CYP2D6 had a 90% decreased risk of chronic manganism compared with the wild-type (Wt/Wt) (odds ratio =0.10, 95% confidence interval =0.01-0.82). A significant association between the CYP2D6 genotype subgroup and the latency of chronic manganese poisoning was also found. Patients who had homozygous (L/L) or heterozygous (Wt/L) mutant alleles developed manganism an average of 10 years later than those who were homozygous wildtype (Wt/Wt). However, the allele and genotype frequencies of CYP1A1 and NQO1 genes were distributed similarly in cases and controls. In addition, no difference in the frequencies of GSTM1 and GSTT1 null genotypes were observed between cases and controls. The results suggest that CYP2D6L gene polymorphism might influence susceptibility to manganese-induced neurotoxicity. However, because of limited sample size, our results should be validated in large-scale studies.

Zhang ZJ, Zhang XB, Hou G, Sha WW, Reynolds GP. 2002. The increased activity of plasma manganese superoxide dismutase in tardive dyskinesia is unrelated to the Ala-9Val polymorphism. J Psychiatr Res 36(5):317-324.
Abstract: That tardive dyskinesia (TD) may have its origins in free-radical toxicity has stimulated investigations into one enzyme important in the control of oxidative free radicals: superoxide dismutase (SOD). The manganese-containing form of this enzyme (MnSOD) is the major superoxide scavenger in mitochondria; a weak association between a functional genetic polymorphism (Ala-9Val) in the mitochondrial targeting sequence (MTS) of this enzyme and TD has been reported in a Japanese population. We have undertaken to determine both the plasma activity of MnSOD and the association of the Ala-9Val polymorphism in a well-matched series of male Chinese schizophrenic patients with (n = 42) and without (n = 59) TD, and normal male controls (n = 50). MnSOD activity was elevated in the TD subjects over those without TD ($P < 0.05$) and normal controls ($P < 0.05$), an effect that was independent of age, age at first antipsychotic treatment, drug dosage and duration of illness. A significant positive correlation between total AIMS score and MnSOD activity was also observed ($P < 0.0001$). No significant reduction in the frequency of the Ala allele was observed in the TD group (0.14) below non-TD (0.18) or control subjects (0.17); nor was there any relationship

between MnSOD activity and the polymorphism. There was no difference between the mean AIMS scores for the two genotypes (V/V and A/V) in the TD group. We conclude that while we have further evidence of a disturbance in the mechanisms regulating oxidative free radicals in TD, this effect is not under the control of the genetic polymorphism investigated here. (C) 2002 Elsevier Science Ltd. All rights reserved.

Zhang FP, Bi SP, Liu JA, Wang XL, Yang XD, Yang L, Yu Q, Hu J, Bai ZP. 2002. Electrochemical and spectrometric studies on the principle of indirect determination of aluminum using L-dopa as an electroactive complexing ligand. Analytical Letters 35(1):135-152.
Abstract: We have shown previously that the anodic peak current of L-dopa decreases linearly with the increase of the concentration of aluminum (Al). In further studies, we have obtained three optimal experimental conditions for the determination of Al in the pH range from 3.6 to 9.2. Here, we are interested in the studies on the principle of the method. That Al coordinates with L-dopa through the electroactive catecholate donor group is demonstrated by physical techniques such as UV-vis, C-13 NMR, and Raman spectrometries. The invariance of the electrooxidized species in the presence of Al is indicated in the plots of DPV anodic peak potential (E-p) with pH, the plots or cyclic voltammetric anodic peak current (I(p)) with square root of scanning rates, (v(1 2)). and charge transfer coefficient (beta) of L-dopa. The calculated pH values of maximum distributions of 1 : 1. 1 : 2. and 1 : 3 Al-L-dOpa complexes are in good agreement with those three optimum pH values for the determination of Al respectively. Therefore, the principle of the method is that Al binding with L-dopa leads to the reduction of electrooxidatable electroactive site of L-dopa. and then results in the decrease of the DPV anodic peak current. The scientific significance of the principle is that it is expected to be fit for both cases: (1) use or ligands with electroactive donor groups similar to L-dopa. and (2) indirect determinations of electrochemical inert species by electroactive chemicals.

Zecca L, Tampellini D, Gatti A, Crippa R, Eisner M, Sulzer D, Ito S, Fariello R, Gallorini M. 2002. The neuromelanin of human substantia nigra and its interaction with metals. J Neural Transm 109(5-6. Sp. Iss. Si):663-672.
Abstract: Neuromelanin (NM) is a peculiar biochemical component of several neurons in the Substantia Nigra (SN), the target area of the degenerative process in Parkinson Disease (PD). SN NM has peculiarities as to its composition and an impressive capacity of chelating metals, iron in particular, but not exclusively. Gaining insights into the structural and functional characteristics of NM should help understanding the reasons of selective vulnerability of nigral neurons in many parkinsonian conditions. From the present data a protective role of NM can be postulated until the buffering capability toward heavy metals are exhausted. The overloading of NM with iron and other metals in neurons may trigger inflammatory and degenerative processes aggravating the underlying pathological condition.

Zecca L, Berg D, Becker G, Riederer P, Musicco M, Gerlach M. 2002. The in vivo detection of iron and ferritins by transcranial sonography: A new approach for early diagnosis of Parkinson's disease. Mov Disord 17:S181.

Yogev-Falach M, Amit T, Bar-Am O, Weinstock M, Youdim MBH. 2002. The involvement of mitogen-activated protein (MAP) kinase in the regulation of amyloid precursor protein processing by novel cholinesterase inhibitors derived from rasagiline. FASEB J 16(10).
Abstract: Two novel neuroprotective cholinesterase (ChE) inhibitors, TV3326, (N-propargyl-(3R) aminoindan-5-yl)-ethyl methyl carbamate, and TV3279, (N-propargyl-(3S) aminoindan-5-yl)ethyl methyl carbamate, were derived from rasagiline for the treatment of Alzheimer's disease (AD). TV3326 also inhibits monoamine oxidase (MAO)-A and -B, whereas its S-isomer, TV3279, lacks MAO inhibitory activity. The action of these drugs in the regulation of amyloid precursor protein (APP) processing, using rat PC12 and human SH-SY5Y neuroblastoma cells, was examined. Both isomers stimulated the release of the non-amyloidogenic alpha-secretase

form of soluble APP (sAPP) from these cell lines. The increases in sAPP induced by TV3326 and TV3279, were dose-dependent (0.1-100 muM) and blocked by the hydroxamic acid-based metalloprotease inhibitor, Ro31-9790, suggesting mediation via alpha-secretase activity. Using several signal transduction inhibitors, we identified the involvement of protein kinase C (PKC), mitogen-activated protein (MAP) kinase, and tyrosine kinase-dependent pathways in the enhancement of sAPP release by TV3326 and TV3279. In addition, both drugs directly induced the phosphorylation of p44 and p42 MAP kinase, which was abolished by the specific inhibitors of MAP kinase activation, PD98059 and U0126. These data suggest a novel pharmacological mechanism whereby these ChE inhibitors regulate the secretory processes of APP via activation of the MAP kinase pathway.

Wuts PGM, Gu RL, Northuis JM, Kwan TA, Beck DM, White MJ. 2002. Development of a practical synthesis of sumanirole. Pure Appl Chem 74(8):1359-1368.
Abstract: A new synthesis of sumanirole that is being developed to treat Parkinson's disease is described. The basic skeleton is constructed from 8-hydroxyquinoline and elaborated to the key tricylic intermediate 14. Further elaboration affords a 1,2-amino alcohol, which is converted to an aziridine by a new process. Finally, dissolving metal reduction to open the aziridine and protecting group removal affords sumanirole.

Wolozin B, Golts N. 2002. Iron and Parkinson's disease. Neuroscientist 8(1): 22-32.
Abstract: Multiple studies implicate iron in the pathophysiology of Parkinson's disease (PD). In the brains of patients with PD, iron levels are elevated and the levels of iron-binding proteins are abnormal. Iron has been suspected to contribute to PD because Fe(II) is known to promote oxidative damage. Recent studies suggest that an additional mechanism by which iron might contribute to PD is by inducing aggregation of the alpha-synuclein, which is a protein that accumulates in Lewy bodies in PD.

Whyte KA, Greenfield SA. 2002. Expression of voltage-dependent calcium channels in the embryonic rat midbrain. Developmental Brain Research 139(2):189-197.
Abstract: The diversity of expression of high-voltage activated voltage-dependent calcium channels (VDCC) was investigated with whole-cell voltage-clamp recordings from dissociated embryonic rat ventral mesencephalic cells over a 7-day culture period. Cell phenotype was identified post-recording by fluorescent immunocytochemistry as tyrosine hydroxylase positive (TH+) or glutamic acid decarboxylase positive (GAD +). Both TH+ and GAD+ cells displayed high-threshold calcium (Ca2+) currents activated by depolarisations positive to -60 mV In both cell types, pharmacological dissection using selective VDCC inhibitors, omega-agatoxin IVA (Aga IVA), to-conotoxin GVIA (GVIA) and nifedipine demonstrated the existence of P/Q-, N- and L-type VDCC, respectively. The remaining residual current could be blocked by cadmium. It was found that the contribution to the whole-cell current by the N-type channel was greater in TH+ cells than GAD+ cells at each time point examined, whilst the contribution to the whole-cell current by the L-type channel was greater in GAD+ cells than TH+ cells. However, over the 7-day culture period, the expression of VDCC types in both cell phenotypes changed in a similar fashion, with the contribution to the whole-cell current from the N-type current decreasing, and the contribution from the R-type current increasing. Our data could provide new insights into a range of neurodevelopmental mechanisms related to Ca2+ homeostasis in developing mesencephalic neurons. (C) 2002 Elsevier Science B.V. All rights reserved.

Weber S, Dorman DC, Lash LH, Erikson K, Vrana KE, Aschner M. 2002. Effects of manganese (Mn) on the developing rat brain: Oxidative-stress related endpoints. Neurotoxicology 23(2):169-175.
Abstract: lie evaluated biochemical endpoints related to oxidative stress in brains of neonatal rats exposed to manganese (Mn). Oral Mn chloride

(MnCl2) (0, 25, or 50 mg Mn chloride kg(-1) body weight per day) was given daily to neonatal rats throughout lactation (i.e. front postnatal day (PND) 1 to 21). As previously reported by (J. Appl. Toxicol. 20 (2000) 179), this treatment paradigm results in increased cerebral cortex (CTX) Mn concentrations in PND 21 rats front both Mn treatment groups. High dose Mn exposure also results in increased cerebellar Mn concentrations. This preliminary study determined whether this exposure paradigm also affects cerebrocortical or cerebellar metallothionein (MT) mRNA levels, glutamine synthetase (GS) activity, GS protein levels, as well as total glutathione (GSH) levels. High dose Mn exposure significantly increased ($P < 0.05$) total cerebrocortical GSH without accompanying changes in any of the other measured parameters. Therefore, it is unlikely that high dose Mn exposure is associated with oxidative stress in this experimental paradigm. (C) 2002 Elsevier Science Inc. All rights reserved.

Walther BW. 2002. Treating restless legs syndrome: current pathophysiological concepts and clinical trials. Expert Opinion on Investigational Drugs 11(4): 501-514.
Abstract: Restless legs syndrome is a distinctive clinical syndrome with a prevalence of about 5% in the general population. One of the outstanding characteristics of restless legs syndrome is its extreme responsiveness to dopaminergic agents. Together with the latest pathophysiological and genetic findings, recent epidemiological and clinical data give a new insight into the classification of restless legs syndrome, thus building the theoretical foundation for the development of new pharmacological methods in its treatment. Current efforts within this area focus on establishing dopaminergic substances for therapy. The hypothesis of a disturbed iron metabolism in restless legs syndrome has been revived by recent theoretical considerations. The present review attempts to explain current strategies of treatment for restless legs syndrome in relation to aetiological, genetic and pathophysiological findings.

Van Baren MJ, Van Der Linde HC, Breedveld GJ, Baarends WM, Rizzu P, De Graaff E, Oostra BA, Heutink P. 2002. A double RING-H2 domain in RNF32, a gene expressed during sperm formation. Biochem Biophys Res Commun 292(1): 58-65.
Abstract: The RING domain is a cysteine-rich zinc-binding motif, which is found in a wide variety of proteins, among which are several proto-oncogenes and the gene implicated in autosomal recessive juvenile parkinsonism, Parkin. The domain mediates binding to other proteins, either via their RING domains or other motifs. In several proteins, RING domains are found in combination with other cysteine-rich binding motifs and some proteins contain two RING domains. Recent evidence suggests that RING finger proteins function in the ubiquitin pathway as E3 ligases. A variant of the RING domain is the RING-H2 domain, in which one of the cysteines is replaced by a histidine. We have cloned and characterized a novel gene, RNF32, located on chromosome 7q36. RNF32 is contained in 37 kb of genomic DNA and consists of 9 constitutive and 8 alternatively spliced exons, most of which are alternative first exons. A long and a short transcript of the gene are expressed; the short transcript containing exons 1-4 only. This gene encodes two RING-H2 domains separated by an IQ domain of unknown function. This is the first reported gene with a double RING-H2 domain. In humans, RNF32 overlaps with a processed retroposon located on the opposite strand, C7orf13. RNF32 is specifically expressed in testis and ovary, whereas C7orf13 is testis-specific, suggesting that its expression may be regulated by elements in the RNF32 promoter region. RNF32 is expressed during spermatogenesis, most likely in spermatocytes and/or in spermatids, suggesting a possible role in sperm formation. (C) 2002 Elsevier Science (USA).

Uversky VN, Li J, Bower K, Fink AL. 2002. Synergistic effects of pesticides and metals on the fibrillation of alpha-synuclein: Implications for Parkinson's disease. Neurotoxicology 23(4-5):527-536.
Abstract: Aggregation of alpha-synuclein has been implicated in the formation of proteinaceous inclusions in the brain (Lewy bodies, Lewy

neurites) that are characteristic of neurodegenerative diseases, such as Parkinson's disease (PD) and dementia with Lewy bodies (DLBs). The etiology of PD is unknown, but recent work has shown that except in rare cases, there appears to be no direct genetic basis. However, several studies have implicated environmental factors, especially pesticides and metals. Here we show that certain pesticides and metals induce a conformational change in a-synuclein and directly accelerate the rate of formation of alpha-synuclein fibrils in vitro. In addition, the simultaneous presence of metal and pesticide led to synergistic effects on the rate of fibrillation. We propose a model in which environmental factors in conjunction with genetic susceptibility may form the underlying molecular basis for idiopathic PD. (C) 2002 Elsevier Science Inc. All rights reserved.

Tsuruta Y, Furuta A, Taniguchi N, Yamada T, Kira J, Iwaki T. 2002. Increased expression of manganese superoxide dismutase is associated with that of nitrotyrosine in myopathies with rimmed vacuoles. Acta Neuropathol (Berl) 103(1): 59-65.
Abstract: Oxidative stress has been suggested as one of the pathogenetic mechanisms of inclusion body myositis (IBM). To study the role of antioxidant enzymes in myopathies with rimmed vacuoles, we examined expressions of copper, zinc superoxide dismutase (Cu, Zn-SOD) and manganese superoxide dismutase (Mn-SOD), and the relationship between SODS and other proteins localized in rimmed vacuoles in muscle biopsy specimens from three cases of sporadic IBM and two of distal myopathy with rimmed vacuoles (DMRV) as well as eight control cases of myopathies without rimmed vacuoles. Immunoblot analysis showed distinct protein bands of both SODS in IBM and DMRV using subtype-specific antibodies. Intensities of immunoreactive bands for Mn-SOD in IBM and DMRV were stronger than those in the control cases. Immunohistochemistry disclosed accumulation of both SODS in vacuolated muscle fibers in all cases of IBM and DMRV Immunoreactivity for Mn-SOD was often colocalized with that of nitrotyrosine, cytochrome oxidase tau, and lysosome-associated membrane proteins 2 (LAMP-2) in vacuolated fibers. Some of the Cu, Zn-SOD-positive vacuolated fibers were associated with ubiquitin. The two SODS may have different roles for cell protection, and the expression of Mn-SOD is associated with nitric oxide-induced oxidative damage in myopathies with rimmed vacuoles.

Tomas-Camardiel M, Herrera AJ, Venero JL, Sanchez-Hidalgo MC, Cano J, Machado A. 2002. Differential regulation of glutamic acid decarboxylase mRNA and tyrosine hydroxylase mRNA expression in the aged manganese-treated rats. Molecular Brain Research 103(1-2):116-129.
Abstract: Recent studies have implicated chronic elevated exposures to environmental agents, such as metals (e.g. manganese, Mn) and pesticides, as contributors to neurological disease. Eighteen-month-old rats received intraperitoneal injections of manganese chloride (6 mg Mn/kg/day) or equal volume of saline for 30 days in order to study the effect of manganese on the dopamine- and GABA-neurons. The structures studied were substantia nigra, striatum, ventral tegmental area, nucleus accumbens and globus pallidus. First, we studied the enzymatic activity of mitochondrial complex 11 succinate dehydrogenase (SDH). We found an overall decrease of SDH in the different brain areas analyzed. We then studied the mRNA levels for tyrosine hydroxylase (TH) and the dopamine transporter (DAT) by in situ hybridization. TH mRNA but not DAT mRNA was significantly induced in substantia nigra and ventral tegmental area following Mn treatment. Correspondingly, TH immunoreactivity was increased in substantia nigra and ventral tegmental area. Manganese treatment significantly decreased GAD mRNA levels in individual GABAergic neurons in globus pallidus but not in striatum. We also quantified the density of glial fibrillary acidic protein (GFAP)-labeled astrocytes and OX-42 positive cells. Reactive gliosis in response to Mn treatment occurred only in striatum and substantia nigra and the morphology of the astrocytes was different than in control animals. These results suggest that the nigrostriatal system could be specifically damaged by manganese toxicity. Thus, changes produced by manganese treatment on 18-month-old rats

could play a role in the etiology of Parkinson's disease. (C) 2002 Elsevier Science B.V. All rights reserved.

Thomas T, Bhavnani BR, Thomas P. 2002. Inhibition of human LDL oxidation by the neuroprotective drug I-deprenyl. Neurol Res 24(2. Sp. Iss. Si): 169-173.
Abstract: L-deprenyl (Selegiline) used in the treatment of Parkinson's and Alzheimer's disease also enhances longevity. Oxidized low density lipoprotein promotes atherosclerosis and is toxic to both vascular and neural tissue. The reported association between vascular dysfunction and neurodegenerative diseases prompted us to investigate the effect of l-deprenyl, a MAO-B inhibitor, on low density lipoprotein (LDL) oxidation. LDL was isolated from freshly collected blood and the kinetics of copper induced oxidation of LDL was monitored continuously by spectrophotometry. Oral administration (10mg) or in vitro (2.8 to 84 muM) addition of l-deprenyl inhibited oxidation of LDL isolated from healthy men and post-menopausal women. This is the first report demonstrating that the antioxidant action of l-deprenyl may be anti-atherogenic and cardioprotective. Such an action could contribute to reported extension of life span associated with long-term administration of the drug. In conjunction with inhibition of LDL oxidation, l-deprenyl is unique in that it demonstrates protective effects on both vascular and neuronal tissue. Prophylactic use of low doses of l-deprenyl may accord protection against vascular and neurodegenerative diseases associated with aging.

Takeda A, Takatsuka K, Sotogaku N, Oku N. 2002. Influence of iron-saturation of plasma transferrin in iron distribution in the brain. Neurochem Int 41(4): 223-228.
Abstract: Based on the evidence that iron distribution in the peripheral tissues is changed by iron-saturation of plasma transferrin, the influence of iron-saturation of plasma transferrin in iron delivery to the brain was examined. Mouse plasma was pre-incubated with ferric chloride in citrate buffer to saturate transferrin and then incubated with (FeCl3)-Fe-59. Peak retention time of Fe-59 was transferred from the retention time of transferrin to that of mercaptalbumin, suggesting that iron may bind to albumin in the plasma in the case of iron-saturation of transferrin. When mice were intravenously injected with ferric chloride in citrate buffer 10 min before intravenous injection of (FeCl3)-Fe-59, Fe-59 concentration in the plasma was remarkably low. Fe-59 concentration in the liver of iron-loaded mice was four times higher than in control, while Fe-59 concentration in the brain of iron-loaded mice was approximately 40% of that of control mice. Twenty-four hours after intravenous injection of (FeCl3)-Fe-59, brain autoradiograms also showed that Fe-59 concentrations in the brain of iron-loaded mice were approximately 40-50% of those of control mice in all brain regions tested except the choroid plexus, in which Fe-59 concentration was equal. These results suggest that the fraction of non-transferrin-bound iron is engulfed by the liver, resulting in the reduction of iron available for iron delivery to the brain in iron-loaded mice. Transferrin-bound iron may be responsible for the fraction of iron in circulation that enters the brain. (C) 2002 Elsevier Science Ltd. All rights reserved.

Tabner BJ, Turnbull S, El-Agnaf OMA, Allsop D. 2002. Formation of hydrogen peroxide and hydroxyl radicals from A beta and alpha-synuclein as a possible mechanism of cell death in Alzheimer's disease and Parkinson's disease. Free Radic Biol Med 32(11):1076-1083.
Abstract: The formation of extracellular or intracellular deposits of amyloid-like protein fibrils is a prominent pathological feature of many different neurodegenerative diseases, including Alzheimer's disease (AD) and Parkinson's disease (PD). In AD, the beta-amyloid peptide (Abeta) accumulates mainly extracellularly at the center or senile plaques, whereas, in PD, the a-synuclein protein accumulates within neurons inside the Lewy bodies and Lewy neurites. We have shown recently that solutions of Abeta 1-40, Abeta 1-42, Abeta 25-35, alpha-synucloin and non-Abeta component (NAC: residues 61-95 of alpha-synuclein) all liberate hydroxyl

radicals upon incubation in vitro followed by the addition of small amounts of Fe(II). These hydroxyl radicals were readily detected by means of electron spin resonance spectroscopy, employing 5,5-dimethyl-1-pyrroline N-oxide (DMPO) as a spin trapping agent, Hydroxyl radical formation was inhibited by the inclusion of catalase or metal-chelators during Abeta or alpha-synuclein incubation. Our results suggest that hydrogen peroxide accumulates during the incubation of Abeta or alpha-synuclein, by a metal-dependent mechanism, and that this is subsequently converted to hydroxyl radicals, on addition of Fe (11), by Fenton's reaction. Consequently, one of the fundamental molecular mechanisms underlying the pathogenesis of cell death in AD and PD, and possibly other neurodegenerative or amyloid diseases, could be the direct production of hydrogen peroxide during formation of the abnormal protein aggregates. (C) 2002 Elsevier Science Inc.

Surh YL, Jung YL, Jang LH, Lee JS, Yoon HR. 2002. Iron enhancement of oxidative DNA damage and neuronal cell death induced by salsolinol. Journal of Toxicology and Environmental Health-Part a 65(5-6):473-488.
Abstract: A group of naturally occurring isoquinoline alkaloids have been detected in certain regions of mammalian brain. One such compound is salsolinol (SAL; 1-methyl-6,7-dihydroxy-1,2,3,4-tetrahydroisoquinoline). This endogenous isoquinoline derivative has been considered to be implicated in the pathophysiology of chronic alcoholism and Parkinsonism. The present study deals with the DNA strand scission induced by SAL in the presence of iron, Incubation of phiX174 DNA with SAL and ferric ion led to conversion of the supercoiled DNA to open circular and linear forms, which was inhibited by the iron chelator deferoxamine, catalase, and scavengers of reactive oxygen species. SAL in combination with Fe(III) also produced 8-hydroxydeoxyguanosine in calf thymus DNA. Exposure of PC12 cells to SAL produced concentration-dependent reduction in viability, which was exacerbated by iron and ameliorated by deferoxamine.

Sipe JC, Lee P, Beutler E. 2002. Brain iron metabolism and neurodegenerative disorders. Dev Neurosci 24(2-3):188-196.
Abstract: Iron, an essential element for central nervous system (CNS) function, has frequently been found to accumulate in brain regions that undergo degeneration in neurological diseases such as Alzheimer disease, Parkinson disease, Friedreich ataxia and other disorders. However, the precise role of iron in the cause of many neurodegenerative diseases is unclear. To assist in understanding the potential importance of iron in CNS disease, this review summarizes the present knowledge in the areas of CNS iron metabolism, homeostasis and disregulation of iron balance caused by mutations in genes encoding proteins involved in iron transport, storage and metabolism. This review encompasses neurodegenerative disorders associated with both iron overload and deficiency to highlight areas where iron misregulation is likely to be important in the pathophysiology of several human brain diseases. Copyright (C) 2002 S. Karger AG, Basel.

Sharpe MA, Ollosson R, Stewart VC, Clark JB. 2002. Oxidation of nitric oxide by oxomanganese-salen complexes: a new mechanism for cellular protection by superoxide dismutase/catalase mimetics. Biochem J 366:97-107.
Abstract: Manganese-salen complexes (Mn-Salen), including EUK-8 [manganese N,N'-bis(salicylidene)ethylenediamine chloride] and EUK-134 [manganese 3-methoxy N,N'-bis(salicylidene)ethylene-diamine chloride], have been reported to possess combined superoxide dismutase (SOD) and catalase mimetic functions. Because of this SOD/catalase mimicry, EUK-8 and EUK-134 have been investigated as possible therapeutic agents in neurological disorders resulting from oxidative stress, including Alzheimer's disease, Parkinson's disease, stroke and multiple sclerosis. These actions have been explained by the ability of the Mn-Salen to remove deleterious superoxide (O-2(-)) and H2O2. However, in addition to oxidative stress, cells in models for neurodegenerative diseases may also be subjected to damage from reactive nitrogen oxides (nitrosative stress), resulting from elevated levels of NO and sister compounds, including peroxynitrite

(ONOO-). We have been examining the interaction of EUK-8 and EUK- 134 with NO and ONOO-. We find that in the presence of a per-species (H2O2, ONOO-, peracetate and persulphate), the Mn-Salen complexes are oxidized to the corresponding oxo-species (oxoMn-Salen). OxoMn-Salens are potent oxidants, and we demonstrate that they can rapidly oxidize NO to NO, and also oxidize nitrite (NO2- to nitrate (NO2-). Thus these Mn-Salens have the potential to ameliorate cellular damage caused by both oxidative and nitrosative stresses, by the catalytic breakdown of O-2(-), H2O2, ONOO- and NO to benign species: O-2(-), H2O, NO2- and NO3-.

Seth K, Agrawal AK, Date I, Seth PK. 2002. The role of dopamine in manganese-induced oxidative injury in rat pheochromocytoma cells. Human & Experimental Toxicology 21(3):165-170.
Abstract: Reactive dopamine (DA) metabolites have been implicated in both Parkinson's disease and manganese (Mn) neurotoxicity. Rat PC12 and genetically modified PC12 (PC12M) cells capable of producing higher DA content, on exposure to MnCl2 (10(-6) M) for 72 hours, exhibited a significant decrease in glutathione content. Activity of antioxidant enzyme catalase was also inhibited following 24- and 72-hour MnCl2 exposure. MnCl2 caused a concentration-dependent (10(-7) to 10(-3) M) loss in mitochondrial activity after 24 and 72 hours and an impaired DNA synthesis after 72 hours with changes being more marked in PC12M cells. The results indicate that the free-radical-mediated toxicity of Mn at cellular level involves down-regulation of antioxidants in normal and DA-rich PC12 cells. PC12M cells appeared to be more sensitive than PC12 cells.

Sangchot P, Sharma S, Chetsawang B, Porter J, Govitrapong P, Ebadi M. 2002. Deferoxamine attenuates iron-induced oxidative stress and prevents mitochondrial aggregation and alpha-synuclein translocation in SK-N-SH cells in culture. Dev Neurosci 24(2-3):143-153.
Abstract: One of the defining characteristics of neurodegenerative diseases, including Parkinson's disease, is an abnormal accumulation of iron in the affected brain areas. By using SK-N-SH, a dopaminergic cell line, we have found that iron (100-250 muM FeSO4) decreased cell viability, increased lipid peroxidation, and the said effects were blocked by deferoxamine (DFO: 10 muM). Furthermore, DFO, in the absence of iron, enhanced the level of adenosine triphosphate (ATP), but caused chromatin condensation and cell death. Morphological studies revealed that iron (50-100 muM) altered mitochondrial morphology, disrupted nuclear membrane, and translocated a-synuclein from perinuclear region into the disrupted nucleus. The results of these studies suggest that DFO is able to block and attenuate iron-mediated oxidative stress. However, in the absence of excess iron, DFO itself may have deleterious effects on the morphology and hence integrity of dopaminergic neurons. Copyright (C) 2002 S. Karger AG, Basel.

Ryan RWJ, Post JI, Solc M, Hodson PV, Ross GM. 2002. Catecholaminergic neuronal degeneration in rainbow trout assessed by skin color change: A model system for identification of environmental risk factors . Neurotoxicology 23(4-5):545-551.
Abstract: Genetic, neurochemical, and environmental factors have been implicated in neurodegenerative disease, and a combination of these factors is likely responsible for disease onset and progression. Environmental toxicants implicated in Parkinson's disease include organic compounds, reactive oxygen species, metal ions and others. Exposure to a combination of environmental toxicants may produce a synergistic insult leading to neuronal death, even though levels of individual toxicants may be below detection by conventional methods. Rodent models of toxicant-induced neurodegeneration are hampered by the high resistance of these animals to many environmental toxicants. Extensive literature on aquatic toxicology and the high homology between many human and fish neurotrophic factors make fish a useful model for investigating environmental toxicants and neurodegeneration. Skin color in salmonids is under catecholaminergic control; pigment-containing melanophores aggregate when stimulated, resulting in paling. We demonstrate that

lesions to nerves innervating melanophores prevent aggregation and produce dark skin color. The time course for return of skin color corresponds to neuronal regeneration, a neurotrophin-dependent event. Observations from this model system may be useful for predicting risks associated with environmental toxicants and nervous system integrity, and may have important implications for the identification of risk factors. (C) 2002 Elsevier Science Inc. All rights reserved.

Roth JA, Horbinski C, Higgins D, Lein P, Garrick MD. 2002. Mechanisms of manganese-induced rat pheochromocytoma (PC12) cell death and cell differentiation. Neurotoxicology 23(2):147-157.
Abstract: Mn is a neurotoxin that leads to a syndrome resembling Parkinson's disease after prolonged exposure to high concentrations. Our laboratory has been investigating the mechanism by which Mn induces neuronal cell death. To accomplish this, ire have utilized rat pheochromocytoma (PC12) cells as a model since they possess much of the biochemical machinery associated with dopaminergic neurons. Mn, like nerve growth factor (NGF), can induce neuronal differentiation of PC12 cells but Mn-induced cell differentiation is dependent on its interaction with the cell surface integrin receptors and basement membrane proteins, vitronectin or fibronectin. Similar to NGF Mn-induced neurtite outgrowth is dependent on the phosphorylation and activation of the MAP kinases, ERKI and 2 (p44/42). Unlike NGF, Mn is also cytotoxic having an IC50 value of similar to600 muM. Although many apoptotic signals are turned on by Mn, cell death is caused ultimately by disruption of mitochondrial function leading to loss of ATP. RT-PCR and immunoblotting studies suggest that some uptake of Mn into PC12 cells depends on the divalent metal transporter 1 (DMTI). DMTI exists in trio isoforms resulting from alternate splicing of a single gene product with one of the two mRNA species containing an iron response clement (IRE) motif downstream from the stop codon. The presence of the IRE provides a binding site for the iron response proteins (IRPI and 2); binding of either of these proteins could stabilize DMTI mRNA and would increase expression of the +IRE form of the transporter. Iron and Mn compete for transport into PC12 cells via DMTl, so removal of iron from the culture media enhances Mn toxicity. The two isoforms of DMTI (IRE) are distributed in different subcellular compartments with the (+/-IRE) species selectively present in the nucleus of neuronal and neuronal-like cells. (C) 2002 Elsevier Science Inc. All rights reserved.

Reaney SH, Kwik-Uribe CL, Smith DR. 2002. Manganese oxidation state and its implications for toxicity. Chem Res Toxicol 15(9):1119-1126.
Abstract: Manganese (Mn) is ubiquitous in mammalian systems and is essential for proper development and function, though it can also be toxic at elevated exposures. While essential biologic functions of Mn depend on its oxidation state [e.g., Mn(II), Mn(III)], little is known about how the oxidation state of elevated Mn exposures affect cellular uptake, and function/toxicity. Here we report the dynamics of EPR measurable Mn(II) in fresh human plasma and cultured PC12 cell lysates as a function of exposure to either manganese(II) chloride or manganese(III) pyrophosphate, and the effects of exposure to Mn(II) versus Mn(III) on total cellular aconitase activity and cellular Mn uptake. The results indicate that Mn(II) or Mn(III) added in vitro to fresh human plasma or cell lysates yielded similar amounts of EPR measurable Mn(II). In contrast, Mn added as Mn(III) was significantly more effective in inhibiting total cellular aconitase activity, and intact PC 12 cells accumulated significantly more Mn when exposures occurred as Mn(III)., Collectively, these data reflect the dynamic nature of Mn speciation in simple biological systems, and the importance of Mn oxidation/speciation state in mediating potential cellular toxicity. This study supports concern over increased environmental exposures to Mn in different oxidation states [Mn(II), Mn(III), and Mn(IV)] that may arise from combustion products of. the gasoline antiknock additive methycyclopentadienyl manganese tricarbonyl (MMT).

Rauhala P, Andoh T, Yeh K, Chiueh CC. 2002. Contradictory Effects of Sodium

Nitroprusside and S-Nitroso-N-Acetylpenicillamine on Oxidative Stress in Brain Dopamine Neurons in VivoVolume 962. p 60-72. Nitric Oxide: Novel Actions, Deleterious Effects and Clinical Potential: Annals of the New York Academy of Sciences.
Abstract: To investigate whether nitric oxide ((NO)-N-.) is neurotoxic or neuroprotective in the brain, we compared the in vivo role of S-nitroso-N-acetyl-penicillamine (SNAP) with that of sodium nitroprusside (SNP) on ferrous citrate-induced oxidative stress and neuronal loss in the rat nigrostriatal dopaminergic system. It is known that light irradiation releases (NO)-N-. from its donor compounds; these irradiated (NO)-N-. donors were used as sham controls in this study. Intranigral infusion of ferrous citrate (4.2 nmol) into the rat midbrain substantia nigra compacta area caused acute lipid peroxidation in the substantia nigra and chronic dopamine depletion in the caudate nucleus. Coinfusion of freshly prepared SNAP (0-8.4 nmol) or (NO)-N-. (about 2 nmol), but not SNP, rescued iron-induced dopamine depletion in the rat brain in vivo. In fact, SNP produced prooxidative effects similar to ferrous citrate both in vivo and in vitro, since SNP is a redox iron complex. Consistently, (NO)-N-. and SNAP inhibited, whereas SNP potentiated, (OH)-O-. generation and lipid peroxidation evoked by ferrous citrate in vitro. We previously reported that freshly prepared, but not irradiated, S-nitroso-L-glutathione (GSNO) protected brain dopamine neurons against oxidative stress in vivo. As well as these antioxidative properties, our recent reports (see Ref. 1) indicate that (NO)-N-./GSNO activated guanylyl cyclase, increased cGMP and that could lead to PKG-mediated expression of MnSOD, Bcl-2, and thioredoxin for preconditioning neuroprotection against 1-methyl-4-phenylpyridinium (MPP +).(1) In conclusion, (NO)-N-. and S-nitrosothiols (e.g., GSNO and SNAP) can scavenge reactive oxygen species and activate the heme moiety of guanylyl cyclase, resulting in protection of brain dopamine neurons through both antioxidative and antiapoptotic mechanisms.

Ramesh GT, Ghosh D, Gunasekar PG. 2002. Activation of early signaling transcription factor, NF-kappa B following low-level manganese exposure. Toxicol Lett 136(2):151-158.
Abstract: Occupational and environmental exposure to manganese (Mn2+) is an increasing problem. It manifests neuronal degeneration characterized by dyskinesia resembling Parkinson's disease. The study was performed to test the hypotheses whether exposure to Mn2+ alters cellular physiology and promotes intracellular signaling mechanism in dopaminergic neuronal cell line. Since transcription factors have been shown to play an essential role in the control of cellular proliferation and survival, catecholaminergic rich pheochromocytoma (PC12) cells were used to measure changes in the DNA binding activities of nuclear factor kappa B (NF-kappaB) by electrophoretic mobility shift assay (EMSA) following Mn2+ (0.1-10 muM) exposure. Cells that were exposed to Mn2+ produced five-fold-activation of transcription factor NF-kappaB DNA binding activity. This remarkable increase was seen within 30-60 min period of Mn2+ exposure. Activation of NF-kappaB DNA binding activity by Mn2+ at 1.0 muM correlated with proteolytic degradation of the inhibitory subunit IkappaBalpha as evidenced in cytosol. Additional experiments on NF-kappaB reporter gene assay also showed increased NF-kappaB gene expression at 1.0 and 5.0 muM Mn2+ and this was completely blocked in the presence of NF-kappaB translocation inhibitor, IkappaBalpha-DN supporting that NF-kappaB induction occurred during Mn2+ exposure. In addition, Mn2+ exposure to PC 12 cells led to activation of signal responsive mitogen activated proteinexposure. In addition, Mn2+ exposure to PC 12 cells led to activation of signal responsive mitogen activated protein kinase kinase (MAPKK). These results suggest that Mn2+ at a low dose appears to induce the expression of immediate early gene, NF-kappaB through MAPKK by a mechanism in which IKBoc phosphorylation may be involved.) (C) 2002 Elsevier Science Ireland Ltd. All rights reserved.

Rachek LI, Grishko VI, Musiyenko SI, Kelley MR, Ledoux SP, Wilson GL. 2002. Conditional targeting of the DNA repair enzyme hOGG1 into mitochondria. J Biol Chem 277(47):44932-44937.

Abstract: Oxidative damage to mitochondrial DNA (mtDNA) has been suggested to be a key factor in the etiologies of many diseases and in the normal process of aging. Although the presence of a repair system to remove this damage has been demonstrated, the mechanisms involved in this repair have not been well defined. In an effort to better understand the physiological role of recombinant 8-oxoguanine DNA glycosylase/ apuriniclyase (OGG1) in mtDNA repair, we constructed an expression vector containing the gene for OGG1 downstream of the mitochondrial localization sequence from manganese-superoxide dismutase. This gene construct was placed under the control of a tetracycline-regulated promoter. Transfected cells that conditionally expressed OGG1 in the absence of the tetracycline analogue doxycycline and targeted this recombinant protein to mitochondria were generated. Western blots of mitochondrial extracts from vector- and OGG1-transfected clones with and without doxycycline revealed that removal of doxycycline for 4 days caused an approximate 8-fold increase in the amount of OGG1 protein in mitochondria. Enzyme activity assays and DNA repair studies showed that the doxycycline-dependent recombinant OGG1 is functional. Functional studies revealed that cells containing recombinant OGG1 were more proficient at repairing oxidative damage in their mtDNA, and this increased repair led to increased cellular survival following oxidative stress.

Racette BA, Antenor J, Kotagal V, Videen T, Moerlein S, Goldman J. 2002. [F-18] FDOPA PET and clinical features in a Parkinson's disease patient with manganese exposure. Mov Disord 17:S108.

Prikhojan A, Brannan T, Yahr MD. 2002. Intrastriatal iron perfusion releases dopamine: an in-vivo microdialysis study. J Neural Transm 109(5-6. Sp. Iss. Si):645-649.
Abstract: We perfused iron as FeCl3 directly into the striatum of normal rats and used the in vivo microdialysis technique to monitor striatal levels of dopamine (DA). KCl was perfused to assess the functional integrity of the DA receptors at the end of each dialysis experiment. Cu+2 (as CuSO4) and Cl- (as NaCl) were perfused to compare the effects of Fe+3 to that of other heavy metal and donors of Cl- anion. Perfusion of FeCl3 (1 mM for 15 min) produced a 250% increase in striatal levels of DA. Perfusion of CuSO4 (1 mM for 15 min) or NaCl (10 mM for 15 min) did not affect striatal DA levels. There was a significant increase in DA levels with KCl stimulation (56 mM for 15 min) after perfusion with FeCl3, We conclude that iron releases DA from striatal nerve endings without the immediate destruction of the DA terminals. The implications of chronic release of dopamine as a cause of dopaminergic cell death are discussed.

Ponzoni S, Gaziri LCJ, Britto LRG, Barreto WJ, Blum D. 2002. Clearance of manganese from the rat substantia nigra following intra-nigral microinjections. Neurosci Lett 328(2):170-174.
Abstract: Chronic exposure to manganese (Mn) positively correlates with the occurrence of Parkinsonism but little is known about mechanisms of its neurotoxicity. In the present study, we determined the clearance of Mn from rat substantia nigra after its nigral injection and correlated it with the establishment of apomorphine-induced rotational behaviour and loss of striatal tyrosine hydroxylase (TH) immunoreactivity. Our results suggest that Mn is slowly cleared from the substantia nigra, following a first-order kinetics with a t(1/2) of 3 days. Appearance of apomorphine-induced rotational behaviour and loss of TH immunoreactivity within the striatum follows metal clearance were both detected 24 hours after intra-nigral Mn microinjection and maximal 72 hours after injection. The present data suggest that the cellular mechanisms induced by Mn and leading to dopaminergic cell death, occurred shortly after its injection and that the metal concentration needs to reach a threshold value to induce neurotoxic effects. This would indicate that nigral damages are a direct consequence of Mn accumulation. (C) 2002 Elsevier Science Ireland Ltd. All rights reserved.

Pifl C, Kattinger A, Reither H, Hornykiewicz O. 2002. Cellular effects of dopamine

- beyond oxidative mechanisms. Parkinsonism & Related Disorders 8(6): 433-437.
Abstract: Cell cycle blockers inhibit growth in dividing cells, but promote survival of differentiated cells, including neurons. Low micromolar dopamine profoundly inhibited cell growth in dopamine transporter transfected SK-N-MC neuroblastoma cells by cell cycle arrest at G(1). This effect was independent of oxy radical formation, antagonized by transporter block, abolished by FeCl3 and mimicked by the iron chelator deferoxamine. We propose that dopamine inhibits cell growth by its ability to chelate intracellular iron. This novel biological action unrelated to neurotransmitter receptors, second messengers or oxidative stress, observed in human neuroblastoma cells of striatal origin, may be important for cell differentiation during neurodevelopment and survival of differentiated dopamine (nigral) neurons. (C) 2002 Published by Elsevier Science Ltd.

Perry G, Sayre LM, Atwood CS, Castellani RJ, Cash AD, Rottkamp CA, Smith MA. 2002. The role of iron and copper in the aetiology of neurodegenerative disorders - Therapeutic implications. Cns Drugs 16(5):339-352.
Abstract: Abnormalities in the metabolism of the transition metals iron and copper have been demonstrated to play a crucial role in the pathogenesis of various neurodegenerative diseases. Metal homeostasis as it pertains to alterations in brain function in neurodegenerative diseases is reviewed in this article in depth, While there is documented evidence for alterations in the homeostasis, redox-activity and localisation of transition metals, it is also important to realise that alterations in specific copper- and iron-containing metalloenzymes appear to play a crucial role in the neurodegenerative process. These changes provide the opportunity to identify pathways where modification of the disease process can occur, potentially offering opportunities for clinical intervention. As understanding of disease aetiology evolves, so do the tools with which diseases are treated. In this article, we examine not only the possible mechanism of disease but also how pharmaceuticals may intervene, from direct and indirect antioxidant therapy to strategies involving gene therapy.

Pattison DI, Dean RT, Davies MJ. 2002. Oxidation of DNA, proteins and lipids by DOPA, protein-bound DOPA, and related catechol(amine)s. Toxicology 177 (1):23-37.
Abstract: Incubation of free 3,4-dihydroxyphenylalanine (DOPA), protein-bound DOPA (PB-DOPA) and related catechols with DNA, proteins and lipids has been shown to result in oxidative damage to the target molecule. This article reviews these reactions with particular emphasis on those that occur in the presence of molecular O-2 and redox-active metal ions (e.g. Fe3+ Cu2+, Cr6+), which are known to increase the rate of DOPA oxidation. The majority of oxidative damage appears to be mediated by reactive oxygen species (ROS) such as superoxide and HO. radicals, though other DOPA oxidation products, including semiquinone radicals, quinones, and metal ion-DOPA complexes have also been implicated in some cases. Non-radical reactions of DOPA with suitable nucleophiles (e.g. thiol groups) can also result in modification of the target, with this process being particularly prevalent with proteins. The exacerbation of damage observed on addition of H2O2 is in accord with a key role for ROS in many of these reactions. (C) 2002 Elsevier Science Ireland Ltd. All rights reserved.

Patel M, Liang LP. 2002. Mitochondrial aconitase inactivation and free iron in experimental Parkinson's disease. Free Radic Biol Med 33:S435.

Patel BN, Dunn RJ, Jeong SY, Zhu QZ, Julien JP, David S. 2002. Ceruloplasmin regulates iron levels in the CNS and prevents free radical injury. J Neurosci 22(15):6578-6586.
Abstract: Ceruloplasmin is a ferroxidase that oxidizes toxic ferrous iron to its nontoxic ferric form. We have previously reported that a glycosylphosphatidylinositol-anchored form of ceruloplasmin is expressed in the mammalian CNS. To better understand the role of ceruloplasmin in iron homeostasis in the CNS, we generated a ceruloplasmin gene-deficient

(Cp-/-) mouse. Adult Cp-/- mice showed increased iron deposition in several regions of the CNS such as the cerebellum and brainstem. Increased lipid peroxidation was also seen in some CNS regions. Cerebellar cells from neonatal Cp-/- mice were also more susceptible to oxidative stress in vitro. Cp-/- mice showed deficits in motor coordination that were associated with a loss of brainstem dopaminergic neurons. These results indicate that ceruloplasmin plays an important role in maintaining iron homeostasis in the CNS and in protecting the CNS from iron-mediated free radical injury. Therefore, the antioxidant effects of ceruloplasmin could have important implications for various neurodegenerative diseases such as Parkinson's disease and Alzheimer's disease in which iron deposition is known to occur.

Park JW, Youn YC, Kwon OS, Jang YY, Han ES, Lee CS. 2002. Protective effect of serotonin on 6-hydroxydopamine- and dopamine-induced oxidative damage of brain mitochondria and synaptosomes and PC12 cells. Neurochem Int 40(3):223-233.
Abstract: The present study elucidated the effects of indoleamines (serotonin, melatonin, and tryptophan) on oxidative damage of brain mitochondria and synaptosomes induced either by 6-hydroxydopamine (6-OHDA) or by iron plus ascorbate and on viability loss in dopamine-treated PC12 cells. Serotonin (1-100 muM), melatonin (100 muM), and antioxidant enzymes attenuated the effects of 6-OHDA, iron plus ascorbate, or 1-methyl-4-phenylpyridinium on mitochondrial swelling and membrane potential formation. Serotonin and melatonin decreased the attenuation of synaptosomal Ca2+ uptake induced by either 6-OHDA alone or iron plus ascorbate. Serotonin and melatonin inhibited the production of reactive oxygen species, formation of malondialdehyde and carbonyls, and thiol oxidation in mitochondria and synaptosomes. and decreased degradation of 2-deoxy-D-ribose. Unlike serotonin, melatonin did not reduce the iron plus ascorbate-induced thiol oxidation. Tryptophan decreased thiol oxidation and 2-deoxy-D-ribose degradation but did not inhibit the production of reactive oxygen species and formation of oxidation products in the brain tissues. Serotonin and melatonin attenuated the dopamine-induced viability loss, including apoptosis, in PC12 cells. The results suggest that serotonin may attenuate the oxidative damage of mitochondria and synaptosomes and the dopamine-induced viability loss in PC12 cells by a decomposing action on reactive oxygen species and inhibition of thiol oxidation and shows the effect comparable to melatonin. Serotonin may show a prominent protective effect on the iron-mediated neuronal damage. (C) 2002 Elsevier Science Ltd. All rights reserved.

Oshiro S, Nozawa K, Hori M, Zhang C, Hashimoto Y, Kitajima S, Kawamura K. 2002. Modulation of iron regulatory protein-1 by various metals. Biochem Biophys Res Commun 290(1):213-218.
Abstract: Iron regulatory protein-1 (IRP-1) is known as a cytosolic aconitase and a central regulator of iron (Fe) homeostasis. IRP-1 regulates the expression of Fe metabolism-related proteins by interacting with the Fe-responsive element (IRE) in the untranslated regions of mRNAs of these proteins. However, it is less known whether IRP-1 modulates various non-Fe metals. In the present study, we showed that treatment of homogenously purified IRP-1 with non-Fe metals decreased the affinity to IRE in RNA band shift assays and increased aconitase activity. Non-Fe metals also inhibited Fe-55 incorporation into the fourth labile position of the Fe-S cluster of IRP-1. In PLC hepatoma cells, metal loading inactivated binding activity and activated enzyme activity. It also suppressed transferrin receptor mRNA expression in the cells. These results suggest that various non-Fe metals modulate IRP-1 by conversion of the 3Fe-4S apo-form to a [1 non-Fe metal + 3Fe]-4Fe holo-form. (C) 2002 Elsevier Science.

Ono K, Komai K, Yamada M. 2002. Myoclonic involuntary movement associated with chronic manganese poisoning. J Neurol Sci 199(1-2):93-96.
Abstract: We report a 17-year-old man showing myoclonic involuntary movement (IVM) associated with chronic manganese (Mn) poisoning. The

patient, a welder, showed myoclonic IVM mainly in the right upper and lower extremities, elevated levels of Mn in the blood and hair and high-intensity signals in the globus pallidus on T1-weighted MR images. Chelation therapy resulted in improvement of the myoclonic IVM and MRI abnormalities. This is the first report of Mn poisoning characterized by myoclonic IVM without parkinsonism. (C) 2002 Elsevier Science B.V. All rights reserved.

Olivieri G, Novakovic M, Savaskan E, Meier F, Baysang G, Brockhaus M, Muller-Spahn F. 2002. The effects of beta-estradiol on SHSY5Y neuroblastoma cells during heavy metal induced oxidative stress, neurotoxicity and beta-amyloid secretion. Neuroscience 113(4):849-855.
Abstract: The role of estrogen as a neurotrophic/neuroprotective agent in neurodegenerative diseases such as Alzheimer's and Parkinson's diseases is increasingly being shown. In this study we examine the neuroprotective effects of beta-estradiol on SHSY5Y neuroblastorna cells which have been exposed to the heavy metals cobalt and mercury. The results show that cobalt and mercury are able to induce oxidative stress and cell cytotoxicity and increase the secretion of beta-amyloid 1-40 and 1-42. These deleterious effects are reversed by the pretreatment of cells with beta-estradiol. It is further shown that beta-estradiol exerts its neuroprotective action through mechanisms which reduce oxidative stress and reduce beta-amyloid secretion. Pre-treatment of the cells with cc-estradiol did not alleviate the toxic effects of the heavy metals. Our results are significant as they contribute to a better understanding of the mode of action of estrogen with relevance to its use in the treatment of neurodegenerative disorders. (C) 2002 IBRO. Published by Elsevier Science Ltd. All rights reserved.

Obata T, Yamanaka Y, Inada T, Kinemuchi H, Oreland L. 2002. In vivo generation of hydroxyl radicals and MPTP-induced dopaminergic neurotoxicity in the striatum. Biogenic Amines 17(1):1-14.
Abstract: Free radical production might make a major contribution at certain stages in the progression of injury to the brain. Oxygen free radical formation has been implicated in lesions caused by 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) and iron. Although MPTP produces a parkinsonian syndrome after its conversion to 1-methyl-4-phenylpyridine (MPP+) by type B monoamine oxidase (MAO-B) in the brain, the etiology of this disease remains obscure. MPP+ is a highly potent dopamine (DA)-releasing agent and DA autoxidation catalyzed by iron and oxidative stress may be involved in the pathogenesis of Parkinson's disease. Histidine, a singlet oxygen (102) scavenger, protects MPP+-induced hydroxyl radical ((OH)-O-.) formation. The inhibitory effect on the susceptibility of LDL oxidation can reduce (OH)-O-. generation. These drugs may be applied as antiparkisonian agents. Further clinical investigation is necessary in future. This finding may be useful in elucidating the actual mechanism of free radical formation in the pathogenesis of neurodegenerative brain disorders, including Parkinson's disease and traumatic brain injuries.

Obata T, Yamanaka Y. 2002. Iron (III) attenuates hydroxyl radical generation accompanying non-enzymatic oxidation of noradrenaline in the rat heart. Naunyn Schmiedebergs Arch Pharmacol 365(2):158-163.
Abstract: The present study examined the effect of iron (III) on the generation of free hydroxyl radicals (.OH) in the extracellular fluid of rat myocardium. The generation of .OH was assessed by infusing sodium salicylate in Ringer's solution (0.5 nmol/mul per min) directly into the myocardium of the anaesthetised rat through a microdialysis probe and measuring the non-enzymatic reaction product 2,3-dihydroxybenzoic acid (DHBA) trapped in the dialysate. Tyramine increased the level of 2,3-DHBA concentration dependently. However, in the presence of iron (III) (50 muM), the effect of tyramine was abolished. When iron (III) (50 muM) was administered to tyramine (1 mM)pre-treated animals, the tyramine-induced stimulation of noradrenaline did not change, but the level of 2,3-DHBA decreased significantly (n=6, $P<0.05$). When desferrioxamine (DES), a

strong iron (III) chelator, was administered to tyramine (1 mM)-pre-treated animals, a marked increase in 2,3-DHBA formation was seen. Administration of iron (II) to the DES-pre-treated animals increased 2,3-DHBA markedly compared with the iron (II)-only treated group, with a positive linear correlation between iron (II) concentration and .OH trapped as 2,3-DHBA (R-2=0.987). DES can reduce iron (III) and thus markedly increases .OH formation. To examine the effect of iron (III) on ischaemia/reperfusion of the myocardium, the heart was subjected to myocardial ischaemia for 15 min by occlusion of the left anterior descending branch of the coronary artery. On reperfusion, noradrenaline and 2,3-DHBA rose markedly in the heart dialysate. The presence of iron (III) (50 muM) abolished the elevation of 2,3-DHBA. Iron (III) also significantly blunted the rise of serum creatine phosphokinase, an index of myocardial damage. The present study demonstrates that the suppression of .OH formation by iron (III) may play a key role in the cardioprotective effect of iron (III) in the rat heart.

Obata T, Egashira T. 2002. Effect of imipramine on 1-methyl-4-phenylpyridinium ion-induced hydroxyl radical generation in rat striatum. Biochimica Et Biophysica Acta-Molecular Basis of Disease 1588(2):173-178.
Abstract: We examined the effect of imipramine (a tricyclic antidepressant drug) on hydroxyl radical (.OH) generation induced by 1-methyl-4-phenylpyridinium ion (MPP+) in extracellular fluid of rat striatum, using a microdialysis technique. Imipramine enhanced the formation of .OH trapped as 2,3-dihydroxybenzoic acid (DHBA) induced by MPP+ (5 mM). Introduction of imipramine (0.1, 0.5 and 1.0 mM) dose-dependently increased the level of dopamine (DA) release. Concomitantly, imipramine enhanced DA efflux and the level of DHBA induced by MPP+, as compared with MPP+-treated control. When corresponding experiments were performed with reserpinized rats, there were small increases in the levels of DA and nonsignificant increase in the formation of DHBA. When iron (11) was administered to imipramine (I mM)-treated animals, a marked elevation of DHBA was observed, compared with MPP+-only treated animals. A positive linear correlation was observed between iron (11) and DHBA (R-2=0.985) in the dialysate. These results indicate that imipramine enhances generation of .OH induced by MPP+ during enhanced DA overflow. (C) 2002 Elsevier Science B.V. All rights reserved.

Obata T, Aomine M, Inada T, Kinemuchi H. 2002. Nicotine suppresses 1-methyl-4-phenylpyridinium ion-induced hydroxyl radical generation in rat striatum. Neurosci Lett 330(1):122-124.
Abstract: The present study examined the ability of antioxidant effects of nicotine on 1-methyl-4-phenylpyridinium ion (MPP+)-induced hydroxyl radical ((OH)-O-.) formation of rat striatum. Rats were anesthetized and sodium salicylate in Ringer's solution (0.5 nmol/mul per min) was infused through a microdialysis probe to detect the generation of (OH)-O-. as reflected by non-enzymatic formation trapped as 2,3-dihydroxybenzoic acid (DHBA) in the striatum. MPP+ enhanced generation of (OH)-O-. formation trapped as DHBA. However, nicotine (100 muM) significantly suppressed (OH)-O-. formation induced by MPP+. Iron (II) (2, 5 and 10 muM) increased the levels of DHBA in a concentration-dependent manner. However, in the presence of nicotine (100 muM), the effect of nicotine was suppressed. These results suggest that nicotine has a preventive effect on (OH)-O-. generation by MPP+ in rat striatum. (C) 2002 Published by Elsevier Science Ireland Ltd.

Obata T. 2002. Role of hydroxyl radical formation in neurotoxicity as revealed by in vivo free radical trapping. Toxicol Lett 132(2):83-93.
Abstract: Reactive oxygen species have been implicated in dopaminergic toxicity caused by 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) and iron. Although MPTP produces a parkinsonian syndrome after its conversion to 1-methyl-4-phenylpyridine (MPP+) by type B monoamine oxidase (MAO-B) in the brain, the etiology of this disease remains obscure. MPPI is a highly potent dopaminbergic-releasing agents and dopamine (DA) autoxidation catalyzed by iron and oxidative stress may be involved in the

pathogenesis of Parkinson's disease. Neuromelanine synthesis from DA produce highly reactive free radicals. Although the controversy possible neurotoxin and/or neuroprotective roles of nitric oxide (NO) was discussed, NO contributes to oxidative injury to brain neurons in vivo. An environmental estrogen-like chemical also related to MPP+-induced OH generation. This review describes actual mechanism of the free radicals formation by dialysis studies of in vivo free radical trapping in the pathogenesis of neurodegenerative disorders, including in the Parkinson's disease, Alzheimer disease and traumatic brain injuries. (C) 2002 Elsevier Science Ireland Ltd. All rights reserved.

Obata T. 2002. Environmental estrogen-like chemicals and hydroxyl radicals induced by MPTP in the striatum: A review. Neurochem Res 27(5):423-431.
Abstract: Oxygen free radical formation has been implicated in lesions caused by the 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) and iron. Although MPTP produces a parkinsonian syndrome after its conversion to 1-methyl-4-phenylpyridine (MPP+) by type B monoamine oxidase (MAO) in the brain, the etiology of this disease remains obscure. This review focuses on the role of an environmental neurotoxin chemically related to MPP+-induced free radical generation in the pathogenesis of Parkinson's disease. Environmental-like chemicals, such as para-nonylphenol or bisphenol A, significantly stimulated hydroxyl radical (.OH) formation in the striatum. Allopurinol, a xanthine oxidase inhibitor, prevents para-nonylphenol and MPP+-induced .OH generation. Tamoxifen, a synthetic nonsteroidal antiestrogen, suppressed the .OH generation via dopamine efflux induced by MPP+. These results confirm that free radical production might make a major contribution at certain stages in the progression of the injury. Such findings may be useful in elucidating the actual mechanism of free radical formation in the pathogenesis of neurodegenerative brain disorders, including Parkinson's disease and traumatic brain injuries.

Normandin L, Hazell AS. 2002. Manganese neurotoxicity: An update of pathophysiologic mechanisms. Metab Brain Dis 17(4):375-387.
Abstract: The central nervous system, and the basal ganglia in particular, is an important target in manganese neurotoxicity, a disorder producing neurological symptoms similar to that of Parkinson's disease. Increasing evidence suggests that astrocytes are a site of early dysfunction and damage; chronic exposure to manganese leads to selective dopaminergic dysfunction, neuronal loss, and gliosis in basal ganglia structures together with characteristic astrocytic changes known as Alzheimer type II astrocytosis. Astrocytes possess a high affinity, high capacity, specific transport system for manganese facilitating its uptake, and sequestration in mitochondria, leading to a disruption of oxidative phosphorylation. In addition, manganese causes a number of other functional changes in astrocytes including an impairment of glutamate transport, alterations of the glycolytic enzyme glyceraldehyde-3-phosphate dehydrogenase, production of nitric oxide, and increased densities of binding sites for the "peripheral-type" benzodiazepine receptor (a class of receptor predominantly localized to mitochondria of astrocytes and involved in oxidative metabolism, mitochondrial proliferation, and neurosteroid synthesis). Such effects can lead to compromised energy metabolism, resulting in altered cellular morphology, production of reactive oxygen species, and increased extracellular glutamate concentration. These consequences may result in impaired astrocytic-neuronal interactions and play a major role in the pathophysiology of manganese neurotoxicity.

Normandin L, Carrier G, Gardiner PF, Kennedy G, Hazell AS, Mergler D, Butterworth RF, Philippe S, Zayed J. 2002. Assessment of bioaccumulation, neuropathology, and neurobehavior following subchronic (90 days) inhalation in Sprague-Dawley rats exposed to manganese phosphate. Toxicol Appl Pharmacol 183(2):135-145.
Abstract: Methylcyclopentadienyl manganese tricarbonyl (MMT) is an organic manganese (Mn) compound added to unleaded gasoline. It has been suggested that the combustion products of MMT containing Mn, such

as manganese phosphate, could cause neurological symptoms similar to Parkinson's disease in humans. The aim of this work was to investigate the exposure-response relationship of bioaccumulation, neuropathology, and neurobehavior following a subchronic inhalation exposure to manganese phosphate in Sprague-Dawley male rats. Rats were exposed 6 h/day, 5 days/week for 13 consecutive weeks at 30, 300, or 3000 mug/m(3) Mn phosphate and compared to controls. Some rats were implanted with chronic EMG electrodes in the gastrocnemius muscle of the hind limb to assess tremor at the end of Mn exposure. Spontaneous motor activity was measured for 36 h using a computerized auto-track system. Rats were then sacrificed by exsanguination and Mn level in different brain tissues and other organs was determined by instrumental neutron activation analysis. Neuronal cell counts were obtained by assessing the sum of five grid areas for the caudate/putamen and the sum of two adjacent areas for the globus pallidus. Increased manganese concentrations were observed in all tissues of the brain and was dose-dependent in olfactory bulb and caudate/putamen. In fact, beginning with the highest level of exposure (3000 mug/m) and ending with the control group, Mn concentrations in the olfactory bulb were 2.47 vs 1.28 vs 0.77 vs 0.64 ppm ($P < 0.05$) while for the caudate/putamen, Mn concentrations were 1.06 vs 0.73 vs 0.62 vs 0.47 ppm ($P < 0.05$). The Mn concentrations in lung were also dose-dependent (10.30 vs 1.40 vs 0.42 vs 0.17 ppm; $P < 0.05$). No statistical difference was observed for loss of neurons in caudate/putamen and globus pallidus. Locomotor activity assessment and tremor assessment did not reveal in neurobehavioral changes between the groups. Our results reinforce the hypothesis that the olfactory bulb and caudate/putamen are the main brain tissues for Mn accumulation after subchronic inhalation exposure. (C) 2002 Elsevier Science (USA).

Niwa J, Ishigaki S, Hishikawa N, Yamamoto M, Doyu M, Murata S, Tanaka K, Taniguchi N, Sobue G. 2002. Dorfin ubiquitylates mutant SOD1 and prevents mutant SOD1-mediated neurotoxicity. J Biol Chem 277(39): 36793-36798.
Abstract: Amyotrophic lateral sclerosis (ALS) is a progressive paralytic disorder resulting from the degeneration of motor neurons in the cerebral cortex, brainstem, and spinal cord. The cytopathological hallmark in the remaining motor neurons of ALS is the presence of ubiquitylated inclusions consisting of insoluble protein aggregates. In this paper we report that Dorfin, a RING finger-type E3 ubiquitin ligase, is predominantly localized in the inclusion bodies of familial ALS with a copper/zinc superoxide dismutase (SOD1) mutation as well as sporadic AILS. Dorfin physically bound and ubiquitylated various SOD1 mutants derived from familial AILS patients and enhanced their degradation, but it had no effect on the stability of the wild-type SOD1. The overexpression of Dorfin protected against the toxic effects of mutant SOD1 on neural cells and reduced SOD1 inclusions. Our results indicate that Dorfin protects neurons by recognizing and then ubiquitylating mutant SOD1 proteins followed by targeting them for proteasomal degradation.

Nguyen A, Gille G, Moldzio R, Hung ST, Rausch WD. 2002. Synthetic neuromelanin is toxic to dopaminergic cell cultures. J Neural Transm 109 (5-6. Sp. Iss. Si):651-661.
Abstract: In the present study, primary cultures of mesencephalic dopaminergic cells were exposed to synthetic dopamine neuromelanin (NM) for 48 hrs at concentrations of 0, 1, 10, 20, 50 and 100 mug NM/ml medium. Differently prepared synthetic NM with or without incorporated iron and NM oxidatively damaged by hydrogen peroxide were used. All NMs affected cellular structures e.g. as swelling of neural processes, rounding of cells, and occasional inclusion of neuromelanin particles. Cell numbers were uniformly and dose dependently reduced. Exposure to MPP+ and ferric iron led to cytotoxic changes which could be further aggravated by oxidatively damaged NM, suggesting cytotoxicity of soluble compounds of NM in predamaged neurons.

Newman MB, Arendash GW, Shytle RD, Bickford PC, Tighe T, Sanberg PR. 2002.

Nicotine's oxidative and antioxidant properties in CNS. Life Sci 71(24): 2807-2820.
Abstract: Nicotine has been reported to be therapeutic in some patients with certain neurodegenerative diseases and to have neuroprotective effects in the central nervous system. However, nicotine administration may result in oxidative stress by inducing the generation of reactive oxygen species in the periphery and central nervous system. There is also evidence suggesting that nicotine may have antioxidant properties in the central nervous system. The antioxidant properties of nicotine may be intracellular through the activation of the nicotinic receptors or extracellular by acting as a radical scavenger in that it binds to iron. The possibility that nicotine might be used to treat some symptoms of certain neurodegenerative diseases underlies the necessity to determine whether nicotine has pro-oxidant, antioxidant or properties of both. This review discusses the studies that have addressed this issue, the behavioral effects of nicotine, and the possible mechanisms of action that result from nicotine administration or nicotinic receptor activation. (C) 2002 Elsevier Science Inc. All rights reserved.

Nappi AJ, Vass E. 2002. Interactions of iron with reactive intermediates of oxygen and nitrogen. Dev Neurosci 24(2-3):134-142.
Abstract: Iron not only functions as a cofactor for various enzymes, but it is also a source of potentially cytotoxic molecules produced through interactions with certain reactive intermediates of oxygen (ROI) and nitrogen (RNI). Protection from such iron-mediated damage results in large part from homeostatic mechanisms that regulate the sequestration of iron. Perturbations in iron homeostasis can result in an array of adverse cellular manifestations including oxidative and nitrosative stress, enhanced production of free radicals, macromolecular damage, and cell death. This brief review focuses on some of the potentially adverse reactions of iron with ROI and RNI. Copyright (C) 2002 S. Karger AG, Basel.

Nakamichi N, Ohno H, Nakamura Y, Hirai T, Kuramoto N, Yoneda Y. 2002. Blockade by ferrous iron of Ca2+ influx through N-methyl-D-aspartate receptor channels in immature cultured rat cortical neurons. J Neurochem 83(1):1-11.
Abstract: Rat cortical neurons cultured for 3 days in vitro were loaded with the fluorescent indicator fluo-3 for assessment of intracellular free calcium ion (Ca2+) concentrations with the aid of a confocal laser-scanning microscope. In the absence of added MgCl2 , the addition of NMDA induced a rapid but sustained increase in the number of fluorescent neurons in a concentration-dependent manner at a concentration range of 1-100 mum with the increase by KCl being transient. The addition of FeCl2 , but not FeCl3 , markedly inhibited the increase by NMDA in a reversible manner at concentrations of 10-200 mum, without affecting that by KCl. Extensive analyses revealed clear differentiation between inhibitions by ferrous iron and other channel blockers known to date. The inhibition by FeCl2 was completely prevented by the addition of two different iron chelators. Exposure to NMDA alone did not lead to cell death in immature cultured neurons, however, while further addition of FeCl2 invariably induced neuronal cell death 24 h after exposure. These results give support to our previous proposal that NMDA receptor complex may contain a novel site sensitive to blockade by ferrous iron in rat brain.

Muller-Lissner S. 2002. General geriatrics and gastroenterology: constipation and faecal incontinence. Best Practice & Research in Clinical Gastroenterology 16(1):115-133.
Abstract: The incidence of constipation increases with age but no consistent changes of colonic or anorectal motility have been shown in elderly people. Instead, neurological diseases, constipating drugs, bedriddenness and weak straining ability may explain this increased prevalence of constipation. The amount of dietary fibre in the diet may be reduced because of poor chewing ability, Parkinson's disease is accompanied by both slow colonic transit and impaired relaxation of the anal sphincter. Drug-induced constipation is particularly likely with anti-parkinsonism drugs

(either anti-cholinergic or dopaminergic) and also with tricyclic antidepressants, opiates, iron, anti-convulsants and aluminium- or calcium-containing antacids. The prevalence of faecal incontinence is also increased in elderly people. About half of frail bedridden institutionalized patients are incontinent. Anal sphincter pressures tend to be lower, but variables of sensitivity are not. In bedridden people faecal impaction may occur. The ensuing rectal distension leads to relaxation of the internal sphincter and hence to faecal soiling. The condition is often overlooked though correct diagnosis is rather simple, being made with a digital rectal examination.

Moos T. 2002. Brain iron homeostasis. Dan Med Bull 49(4):279-301.

Mondino F, Filippi P, Magliola U, Duca S. 2002. Magnetic resonance relaxometry in Parkinson's disease. Neurological Sciences 23:S87-S88.
Abstract: A central role of iron in the pathogenesis of idiopathic Parkinson's disease (PD), due to its increase in substantia nigra pars compacta dopaminergic neurons and its capacity to enhance production of toxin reactive oxygen radicals, has been discussed for many years. Nuclear magnetic resonance (NMR) relaxation is considered an objective and noninvasive method of measuring regional iron concentrations. By means of this technique we investigated both controls and PD patients.

Mohanakumar KP, Thomas B, Sharma SM, Muralikrishnan D, Chowdhury R, Chiueh CC. 2002. Nitric Oxide - an Antioxidant and NeuroprotectorVolume 962. p 389-401. Nitric Oxide: Novel Actions, Deleterious Effects and Clinical Potential: Annals of the New York Academy of Sciences.
Abstract: Indirect evidence, including neuroprotection against 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP)-neurotoxicity by nitric oxide synthase (NOS) inhibitors and resistance of transgenic animals deficient in NOS, is controversial. We have reviewed evidence in favor of oxidative stress during the development of MPTP-neurotoxicity and the influence of antioxidants, including nitric oxide (NO) and NO donors, on MPTP-induced dopaminergic neurotoxicity. Systemic administration of MPTP causes dose-dependent generation of hydroxyl radicals (OH) in vivo in the striatum in mice; OH scavengers protect dopaminergic neurons from this insult. On the other hand the role of NO in MPTP-neurotoxicity is controversial. Hitherto, no direct evidence for the involvement of NO in MPTP neurotoxicity has been available. MPTP does not affect inducible-NOS mRNA level or its expression in SN or the striatum. Nitroglycerine, a NO donor, can attenuate MPTP-induced dopamine depletion in the striatum by virtue of its OH scavenging action. Several other NO donors have also been shown to scavenge the OH generated, following Fenton chemistry in vitro, and to protect against in vivo dopaminergic neurotoxicity by small mass iron complex formation. This evidence suggests that NO renders protection against MPTP-induced OH-mediated nigrostriatal lesions, acting as an antioxidant.

Milligan SA, Chesson AL. 2002. Restless legs syndrome in the older adult - Diagnosis and management. Drugs & Aging 19(10):741-751.
Abstract: Restless legs syndrome (RLS) is common in the elderly, with an estimated prevalence of 10 to 35% in individuals over 65 years of age. RLS is characterised by paraesthesias and dysaesthesias of the legs, typically occurring in the evening. The symptoms occur at rest and result in motor restlessness; movement often temporarily relieves the symptoms. Patients with poorly controlled RLS may develop related problems including insomnia (due to sleep-onset restlessness or periodic limb movements or related sleep fragmentation) and depression. RLS can be a primary disorder that develops in the young and includes familial cases. Secondary RLS occurs in association with iron-deficiency anaemia, uraemia and polyneuropathies. Typically, RLS is misdiagnosed or undiagnosed for years. In the elderly, both primary and secondary types of the disorder are common. It is thought that RLS represents lower CNS levels of, or reduced responsiveness to, dopamine. The symptoms improve with dopaminergic therapy. Ergotamine dopamine-receptor agonists such as pergolide, and the non-ergotamine dopamine-receptor agonists

pramipexole and ropinirole, are becoming more commonly used to treat RLS. The dopamine precursor levodopa, in combination with carbidopa, is another effective therapeutic agent. An advantage of levodopa is lower cost than non-ergotamine and ergotamine dopamine-receptor agonists. However, the adverse effect of symptom augmentation appears to develop more frequently with levodopa than dopamine-receptor agonists; therefore, levodopa may currently be used somewhat less often as first-line therapy. Patients with painful symptoms may respond favourably to the anticonvulsants gabapentin and carbamazepine. Opioids and hypnosedatives are helpful in selected patients; however, these agents may have troubling adverse effects in the elderly. Correction of iron deficiency improves symptoms in patients with low ferritin levels. Lifestyle modification may also be helpful. Therapy is directed at symptoms, and most symptomatic patients benefit from treatment. It is important to consider RLS in the differential diagnosis of any patient with paraesthesias of the limbs.

Mendez-Alvarez E, Soto-Otero R, Hermida-Ameijeiras A, Lopez-Real AM, Labandeira-Garcia JL. 2002. Effects of aluminum and zinc on the oxidative stress caused by 6-hydroxydopamine autoxidation: relevance for the pathogenesis of Parkinson's disease. Biochimica Et Biophysica Acta-Molecular Basis of Disease 1586(2): 155-168.
Abstract: Aluminum and zinc have been related to the pathogenesis of Parkinson's disease (PD), the former for its neurotoxicity and the latter for its apparent antioxidant properties. 6-Hydroxydopamine (6-OHDA) is an important neurotoxin putatively involved in the pathogenesis of PD, its neurotoxicity often being related to oxidative stress. The potential effect of these metals on the oxidative stress induced by 6-OHDA autoxidation and the potential of ascorbic acid (AA), cysteine, and glutathione to modify this effect were investigated. Both metals, particularly Al3+, induced a significant reduction in (OH)-O-. production by 6-OHDA autoxidation. The combined action of AA and a metal caused a significant and sustained increase in (OH)-O-. generation, particularly with Al3+, while the effect of sulfhydryl reductants was limited to only the first few minutes of the reaction. However, both Al3+ and Zn2+ provoked a decrease in the lipid peroxidation induced by 6-OHDA autoxidation using mitochondrial preparations from rat brain, assessed by TBARS formation. In the presence of AA, only Al3+ induced a significant reduction in lipid peroxidation. After intrastriatal injections of 6-OHDA in rats, tyrosine hydroxylase immunohistochemistry revealed that Al3+ reduces 6-OHDA-induced dopaminergic lesion in the striatum, which corroborates the involvement of lipid peroxidation in 6-OHDA neurotoxicity and appears to discard the participation of this mechanism on PD by Al3+ accumulation. The previously reported antioxidant properties of Zn2+ appear to be related to the induction of Zn2+-containing proteins and not to the metal per se. (C) 2001 Elsevier Science B.V. All rights reserved.

Malecki EA, Cable EE, Isom HC, Connor JR. 2002. The lipophilic iron compound TMH-ferrocene [(3,5,5-trimethylhexanoyl)ferrocene] increases iron concentrations, neuronal L-ferritin, and heme oxygenase in brains of BALB/c mice. Biol Trace Elem Res 86(1):73-84.
Abstract: Mismanagement of intracellular iron is a key pathological feature of many neurodegenerative diseases. Our long-term goal is to use animal models to investigate the mechanisms of iron neurotoxicity and its relationship to neurodegenerative pathologies. The immediate aim of this experiment was to determine regional distribution of iron and cellular distribution of iron storage proteins (L- and H-ferritin) and an oxidative stress marker (heme oxygenase-1) in brains of mice fed the lipophilic iron compound (3,5,5-trimethylhexanoyl) (TMH)-ferrocene. We fed male and female weanling BALB/cj mice diets either deficient in iron (0 mg Fe/kg diet), adequate in iron (35 mg Fe/kg diet; control mice), or adequate in iron and supplemented with 0.1 or 1.0 g TMH-ferrocene/kg diet for 8 wk. Iron concentrations in cerebrum were higher in mice fed 1.0 g TMH-ferrocene/kg diet than in control mice ($p < 0.05$). Liver iron concentrations were eightfold higher in mice fed 1.0 g TMH-ferrocene/kg diet than in

control mice ($P < 0.0001$). L-Ferritin and heme oxygenase-1 expression were elevated in striatum in mice fed 1.0 g TMH-ferrocene/kg diet. We conclude that administration of the lipophilic iron compound TMH-ferrocene leads to subtle perturbations of cellular iron within the brain, potentially representing a model of iron accumulation similar to that seen in various neuropathological conditions.

Lorenzl S, Albers DS, Narr S, Chirichigno J, Beal MF. 2002. Expression of MMP-2, MMP-9, and MMP-1 and their endogenous counterregulators TIMP-1 and TIMP-2 in postmortem brain tissue of Parkinson's disease. Exp Neurol 178 (1):13-20.
Abstract: We investigated the levels and tissue localization of matrix metalloproteinase 2 (MMP-2) and matrix metal. loproteinase 9 (MMP-9) in postmortem brain tissue from Parkinson's disease (PD) and age-matched control cases. Using zymography, we found reduced MMP-2 levels in PD cases in the substantia nigra as compared to controls; levels of MMP-2 were not significantly changed in the cortex and the hippocampus. MMP-9 levels were unchanged in the investigated brain regions. Immunohistochemically, MMP-2 was localized primarily in astrocytes and microglia cells, whereas MMP-9 was predominantly neuronal. Levels of MMP-1, an endogenous tissue inhibitor of MMPs, were significantly elevated in the substantia nigra, but not in the cortex and hippocampus. TIMP-2 levels were unchanged in PD. To investigate whether increased TIMP-1 levels in the substantia nigra might be due to increased MMP-1 expression, we measured MMP-1 levels using Western blots. MMP-1 levels were unchanged in PD cases compared to controls. Together, these data show alterations of MMP-2 and TIMP-1 in the substantia nigra of PD, consistent with the possibility that alterations in MMPs/TIMPs may contribute to disease pathogenesis. (C) 2002 Elsevier Science (USA).

Lindh U, Hudecek R, Danersund A, Eriksson S, Lindvall A. 2002. Removal of dental amalgam and other metal alloys supported by antioxidant therapy alleviates symptoms and improves quality of life in patients with amalgam-associated ill health. Neuroendocrinology Letters 23(5-6):459-482.
Abstract: OBJECTIVES: The purpose of this study was to evaluate treatment of patients suffering from chronic ill health with a multitude of symptoms associated with metal exposure from dental amalgam and other metal alloys. SETTING AND DESIGN: We included 796 patients in a retrospective study using a questionnaire about symptom changes, changes in quality of life as a consequence of treatment and assessment of care taking. METHODS: Treatment of the patients by removal of offending dental metals and concomitant antioxidant therapy was implemented according to the Uppsala model based on a close co-operation between physicians and dentists. RESULTS: More than 70% of the responders, remaining after exclusion of those who had not begun or completed removal, reported substantial recovery and increased quality of life. Comparison with similar studies showed accordance of the main results. Plasma concentrations of mercury before and after treatment supported the metal exposure to be causative for the ill health. MAIN FINDINGS: Treatment according to the Uppsala model proved to be adequate for more than 70% of the patients. Patients with a high probability to respond successfully to current therapy might be detected by symptom profiles before treatment. CONCLUSIONS: The hypothesis that metal exposure from dental amalgam can cause ill health in a susceptible part of the exposed population was supported. Further research is warranted to develop laboratory tests to support identification of the group of patients responding to current therapy as well as to find out causes of problems in the group with no or negative results (250 words).

Lin AMY, Chen CF, Ho LT. 2002. Neuroprotective effect of intermittent hypoxia on iron-induced oxidative injury in rat brain. Exp Neurol 176(2):328-335.
Abstract: The neuroprotective effect of intermittent hypoxia on ferrous citrate (iron)-induced oxidative stress was investigated in the nigrostriatal dopaminergic system of rat brain. Female Wistar rats were subjected to 380 mm Hg in an altitude chamber for 15 h/day for 7, 14, or 28 days. Iron

was locally infused in the substantia nigra of anesthetized rats. Seven days after infusion, lipid peroxidation was elevated in the infused substantia nigra and dopamine content and tyrosine hydroxylase-positive axons were decreased in the ipsilateral striatum in the normoxic rats. Intermittent hypoxic treatment prevented iron-induced oxidative injuries. Induction of the neuroprotection required 2 weeks. Intracerebroventricular infusion of L-buthionine-[S,R]-sulfoximine (L-BSO), which mimicked a reduced antioxidative condition, aggravated iron-induced oxidative injuries. Intermittent hypoxia ameliorated L-BSO-induced augmentation of iron-induced oxidative injuries. Basal GSH (glutathione) content, GSH/GSSG ratio, superoxide dismutase (SOD) and catalase activities in intact substantia nigra were not altered by intermittent hypoxia. Furthermore, intermittent hypoxia attenuated iron-induced reductions in GSH content, GSH/GSSG ratio, and SOD, iron-induced increase in catalase but had no effect on glutathione peroxidase. Our data suggest that intermittent hypoxia may protect the nigrostriatal dopaminergic system from iron-induced oxidative injuries. Moreover, antioxidative defensive systems may partially contribute to the neuroprotection by intermittent hypoxia. (C) 2002 Elsevier Science (USA).

Li YB, Cao ZX. 2002. The neuroprotectant ebselen inhibits oxidative DNA damage induced by dopamine in the presence of copper ions. Neurosci Lett 330(1): 69-73.
Abstract: Ebselen (2-phenyl-1,2-benzisoselenazol-3(2H)-one), a seleno-organic compound with glutathione peroxidase-like activity, has been shown to be protective against brain ischemic injury and Parkinson's disease. This study was undertaken to investigate the protective effects of ebselen on oxidative DNA damage induced by dopamine in the presence of copper ions. Incubation of phiX-174 plasmid DNA with micromolar dopamine in the presence of Cu(II) resulted in a concentration-dependent induction of DNA strand breaks. Both a Cu(II)/Cu(I) redox cycle and H2O2 formation were critically involved in the induction of DNA strand breaks by the dopamine/Cu(II) system. The presence of ebselen at micromolar concentrations led to a marked concentration-dependent inhibition of DNA strand breaks induced by the dopamine/Cu(II) system. Further studies showed that ebselen did not affect either the Cu(II)-mediated oxidation of dopamine to dopamine quinone or the reduction of Cu(II) to Cu(I) by dopamine. Instead, the presence of ebselen resulted in a marked decrease in the levels of H2O2 derived from the Cu(II)-mediated oxidation of dopamine. Taken together, our results demonstrate for the first time that ebselen is able to inhibit the dopamine/Cu(I)-induced oxidative DNA damage, which appears to be attributable to the ability of ebselen to decrease the levels of H2O2 derived from the dopamine/Cu(II) system. Since oxidative DNA damage has been implicated in the pathogenesis of various neurodegenerative diseases, the inhibition of oxidative DNA damage by ebselen may be responsible, at least partially, for its neuroprotective activities observed in both humans and experimental animals. (C) 2002 Elsevier Science Ireland Ltd. All rights reserved.

Levites Y, Youdim MBH, Maor G, Mandel S. 2002. Attenuation of 6-hydroxydopamine (6-OHDA)-induced nuclear factor-kappaB (NF-kappa B) activation and cell death by tea extracts in neuronal cultures. Biochem Pharmacol 63(1):21-29.
Abstract: Antioxidant and anti-inflammatory therapy approaches have been in the focus of attention in the treatment of neurodegenerative Parkinson's and Alzheimer's diseases where oxidative stress has been implicated, Tea extracts have been previously reported to possess radical scavenger, iron chelating and anti-inflammatory properties in a variety of tissues. The purpose of this study was to investigate potential neuroprotective effects of tea extracts and possible signal pathway involved in a neuronal cell model of Parkinson's disease. We demonstrated highly potent antioxidant-radical scavenging activities of green tea (GT) and black tea (BT) extracts on brain mitochondrial membrane fraction. against iron (2.5 muM)-induced lipid peroxidation. Both extracts (0.6-3 muM total polyphenols) were shown to attenuate the neurotoxic action of

6-hydroxydopamine (6-OHDA)-induced neuronal death. 6-OHDA (350 and 50 muM) activated the iron dependent inflammatory redox sensitive nuclear factor-kappaB (NF-kappaB) in rat pheochromocytoma (PC 12) and human neuroblastoma (NB) SH-SY5Y cells, respectively. Immunofluorescence and electromobility shift assays showed increased nuclear translocation and binding activity of NF-kappaB after exposure to 6-OHDA in NB SH-SY5Y cells, with a concomitant disappearance from the cytoplasm. Introduction of GT extract (0.6. 3 muM total polyphenols) before 6-OHDA inhibited both NF-kappaB nuclear translocation and binding activity induced by this toxin in NB SH-SY5Y cells. Neuroprotection was attributed to the potent antioxidant and iron chelating actions of the polyphenolic constituents of tea extracts, preventing nuclear translocation and activation of cell death promoting NF-kappaB. Brain penetrating property of polyphenols may make such compounds an important class of drugs for treatment of neurodegenerative diseases. (C) 2002 Elsevier Science Inc. All rights reserved.

Lee PL, Gelbart T, West C, Halloran C, Sipe JC, Beutler E. 2002. Polymorphisms in iron-responsive binding protein 2 and lack of association with sporadic Parkinson's disease. Mov Disord 17(6):1302-1304.
Abstract: Mice with targeted disruptions in the iron-responsive binding protein 2 (IRP2) gene accumulate iron in distinct regions of the brain and develop neurodegenerative characteristics resembling Parkinson's disease after 6 months of age. To determine whether polymorphisms in IRP2 predispose humans to Parkinson's disease (PD), we sequenced the IRP2 gene of subjects with sporadic PD and normal controls. Three polymorphisms which result in an amino acid change were identified: L159V, F272L, and T560I. The L159V and T560I polymorphisms, identified in an African-American PD subject, were found in the African-American population at an allele frequency of 0.102 (n = 1,236) and 0.111 (n = 1,228) respectively, and were not associated with an increased prevalence of PD. The F272L polymorphism was found in a normal 58-year-old, Caucasian subject whose father had PD, but it was not observed in 38 additional patients with sporadic PD. The F272L polymorphism occurred at an allele frequency of 0.0014 (n = 1,384) in the normal Caucasian population. Additional F272L heterozygous subjects identified in the normal population did not have a family or personal history of PD. We conclude that these IRP2 polymorphisms do not play an important role in the development of sporadic cases of PD. It remains to be determined whether other polymorphisms in IRP2 play a role in familial PD. (C) 2002 Movement Disorder Society.

Leary SC, Hill BC, Lyons CN, Carlson CG, Michaud D, Kraft CS, Ko K, Glerum DM, Moyes CD. 2002. Chronic treatment with azide in situ leads to an irreversible loss of cytochrome c oxidase activity via holoenzyme dissociation. J Biol Chem 277(13):11321-11328.
Abstract: Chronic treatment of cultured cells with very low levels of azide (I-50 < 10 mu(M)) leads to slow (t(1/2) = 6 h), irreversible loss of cytochrome c oxidase (COX) activity. Azide-mediated COX losses were not accompanied by inhibition of other mitochondrial enzymes and were not dependent upon electron flux through oxidative phosphorylation. Although azide treatment also reduced activity (but not content) of both CuZn superoxide dismutase and catalase, a spectrum of pro-oxidants (and anti-oxidants) failed to mimic (or prevent) azide effects, arguing that losses in COX activity were not due to resultant compromises in free radical scavenging. Loss of COX activity was not attributable to reduced rates of mitochondrial protein synthesis or declines in either COX subunit mRNA or protein levels (COX I, II, IV). Co-incubation experiments using copper (CuCl2 Cu-His) and copper chelators (neocuproine, bathocuproine) indicated that azide effects were not mediated by interactions with either Cu-A or Cu-B. In contrast, difference spectroscopy and high performance liquid chromatography analyses demonstrated azide-induced losses in cytochrome aa(3) content although not to the same extent as catalytic activity. Differential azide effects on COX content relative to COX activity were confirmed using a refined inhibition time course in combination with

blue native electrophoresis, and established that holoenzyme dissociation occurs subsequent to losses in catalytic activity. Collectively, these data suggest that COX deficiency can arise through enhanced holoenzyme dissociation, possibly through interactions with the structure or coordination of its heme moieties.

Lazeyras FO, Spahr L, Dupasquier R, Delavelle J, Burkhard P, Hadengue A, Hochstrasser D, Mentha G, Giostra E, Terrier F, Vingerhoets F. 2002. Persistence of mild parkinsonism 4 months after liver transplantation in patients with preoperative minimal hepatic encephalopathy: a study on neuroradiological and blood manganese changes. Transpl Int 15(4): 188-195.
Abstract: Pallidal hyperintensity at magnetic resonance imaging (MRI) correlates to blood manganese (Mn) levels and parkinsonian signs in patients with cirrhosis. Similarly, metabolite changes in the basal ganglia (BG) at proton spectroscopy are related to these neurological signs. The evolution of these abnormalities after liver transplantation (OLT) is incompletely described. We evaluated 14 unselected consecutive patients with cirrhosis (minimal hepatic encephalopathy [HE] n = 8, no HE n = 6) before and 4 months after successful OLT for the evolution of parkinsonism using a validated scale (the United Parkinson's Disease Rating Scale, or UPDRS). Pallidal intensity at MRI, spectroscopic changes in the BG at magnetic resonance spectroscopy (MRS), and whole blood manganese concentrations were measured. After OLT in patients with preoperative minimal HE, the UPDRS scores improved, but mild parkinsonism persisted (16.1 +/- 3.6 to 6.2 +/- 4.8, P < 0.05). Pallidal hyperintensity remained abnormal in 5/8 of cases, but spectroscopic changes normalized in all patients, Blood Mn remained elevated in 4/6 patients. In patients without HE, UPDRS values remained negligible (2.42 +/- 1.5 to 2.5 +/- 1.4). Pallidal hyperintensity normalized in 7/8 patients and spectroscopic changes normalized in all patients. Blood Mn remained elevated in 5/6 patients. Four months after successful OLT, patients with preoperative minimal HE and severe pallidal hyperintensity showed persistent mild parkinsonism. The role of blood manganese determination appears limited in the monitoring of MRI and parkinsonian signs changes after OLT.

Lai BCL, Marion SA, Teschke K, Tsui JKC. 2002. Occupational and environmental risk factors for Parkinson's disease. Parkinsonism & Related Disorders 8 (5):297-309.
Abstract: The etiology of Parkinson's disease (PD) remains obscure. Current research suggests that a variety of occupational and environmental risk factors may be linked to PD. This paper provides an overview of major occupational and environmental factors that have been associated with the development of PD and tries to assess current thinking about these factors and their possible mechanisms of operation. While clear links to rural living, dietary factors, exposure to metals, head injury, and exposure to infectious diseases during childhood have not been established, there is general agreement that smoking and exposure to pesticides affect the probability of developing PD. (C) 2002 Elsevier Science Ltd. All rights reserved.

Kunikowska G, Jenner P. 2002. The distribution of copper, zinc- and manganese-superoxide dismutase, and glutathione peroxidase messenger ribonucleic acid in rat basal ganglia. Biochem Pharmacol 63(6):1159-1164.
Abstract: Oxidative stress may contribute to the progression of Parkinson's disease, and while the status of antioxidant enzymes is thus important, little data on their regional distribution in basal ganglia exist. We now report on the distribution and levels of messenger ribonucleic acid (m-RNA) for the antioxidant enzymes copper, zinc-superoxide dismutase (Cu,Zn-SOD), manganese-superoxide dismutase (Mn-SOD), and glutathione peroxidase in rat basal ganglia using in situ hybridisation histochemistry with complementary deoxyribonucleic acid probes specific for these enzymes. The m-RNA for Cu,Zn-SOD, Mn-SOD. and glutathione peroxidase was expressed throughout basal ganglia. Levels of m-RNA were significantly higher in substantia nigra pars compacta than in all other

regions of basal ganglia for both Cu,Zn-SOD (53-62%, P < 0.001) and Mn-SOD (37-45%, P < 0.05). Mn-SOD m-RNA levels were also significantly higher in SN pars reticulata than in the nucleus accumbens (10%, P < 0,05) and striatum (12%, P < 0,01). In contrast, glutathione peroxidase m-RNA levels were only significantly higher in SN pars compacta when compared with SN pars reticulata (23%, P < 0.05), and in the striatum when compared with the nucleus accumbens (21%, P < 0.05). The data suggest that SN pars compacta may be vulnerable to oxidative stress and thus dependent on the high antioxidant capacity provided by these cytoprotective enzymes. In conclusion, this study demonstrates the relative distribution of antioxidant enzymes in rat basal ganglia and forms the basis for further study in rodent models of Parkinson's disease. (C), 2002 Elsevier Science Inc. All rights reserved.

Kunikowska G, Gallagher I, Glover V, Clow A, Jenner P. 2002. Effects of short- and long-term (-)-deprenyl administration on mRNA for copper, zinc- and manganese-superoxide dismutase and glutathione peroxidase in rat brain. Brain Res 953(1-2):1-11.
Abstract: The effect of short-term (3 weeks, 2 mg/kg day) and long-term (12 and 20 months, 0.5 mg/kg day) administration of (-)-deprenyl on the mRNA expression of three neuroprotective enzymes in subdivisions of rat basal ganglia was investigated. In situ hybridisation histochemistry with oligodeoxynucleotide probes was used to measure levels of copper, zinc superoxide dismutase (Cu,Zn-SOD), manganese superoxide dismutase (Mn-SOD), and glutathione peroxidase (GPX) mRNA. The 3-week administration of (-)-deprenyl caused a significant increase in Cu,Zn-SOD mRNA in the nucleus accumbens (NA) (P<0.05), striatum (CP) (P<0.01), and globus pallidus (GP) (P<0.05), but had no effect on Mn-SOD or GPX mRNA levels throughout basal ganglia. In rats which received (-)-deprenyl for 12 months, there was a significant increase in Mn-SOD mRNA in the NA, CP GP, and substantia nigra (SN) (all P<0.05); there were no changes in either Cu,Zn-SOD or GPX mRNA. Except for the significant increase in Cu,Zn-SOD mRNA in SN pars compacta (SC) (P<0.05), by 20 months there were almost no differences between (-)-deprenyl-treated and age-matched control animals that had received equivalent injections of saline. We conclude that (-)-deprenyl administration can induce mRNA expression for both forms of SOD, but the effects are variable and not sustained over 20 months. (C) 2002 Elsevier Science B.V. All rights reserved.

Kumar A. 2002. Movement disorders in the tropics. Parkinsonism & Related Disorders 9(2):69-75.
Abstract: The spectrum of movement disorders in the tropics is different from that seen in the industrialized nations of the west. This is not surprising given the unique combination of environmental and population characteristics in the tropics. Infections seldom encountered in the west such as tuberculous meningitis, typhoid fever, Japanese encephalitis, malaria, trypanosomiasis or cysticercosis are often seen. in the tropics and with global patterns of travel and immigration these conditions are becoming more common worldwide. Movement disorders associated with these infections, HIV, slow virus and prion disease are discussed. Taking into account the diverse etiologies of movement disorders in the tropics, movement disorders with a nutritional basis such as the infantile tremor syndrome, seasonal ataxia and tropical ataxic neuropathy, and manganese neurotoxicity are also reviewed. Finally, certain special characteristics of ubiquitous disorders such as Parkinson's disease, and disorders with a genetic basis such as Wilson's disease and spinocerebellar degeneration are described. (C) 2002 Elsevier Science Ltd. All rights reserved.

Kress GJ, Dineley KE, Reynolds IJ. 2002. The relationship between intracellular free iron and cell injury in cultured neurons, astrocytes, and oligodendrocytes. J Neurosci 22(14):5848-5855.
Abstract: Iron is an essential element for cells but may also be an important cytotoxin. However, very little is known about iron transport, redox status, or toxicity specifically inside cells. In this study, we exploited the sensitivity of fura-2 to quenching by ferrous iron (Fe2+) to detect

intracellular free iron ([Fe2+](i))in neurons, astrocytes, and oligodendrocytes in primary culture. All cell types exposed to Fe2+ in the presence of the ionophore pyrithione rapidly accumulated Fe2+ to a similar extent. The heavy-metal chelators bipyridyl and N,N,N',N'- tetrakis(2-pyridalmethyl) ethyl-enediamine rapidly reversed the increase in [Fe2+](i), whereas desferrioxamine had little effect. Interestingly, the Fe2+-mediated quenching of fura-2 fluorescence was reversed in a concentration-dependent manner by hydrogen peroxide. This was likely caused by the oxidation of Fe2+ to Fe3+ inside the cell. Acute exposure of cells to Fe2+ was only toxic when the metal was applied together with pyrithione, showing that Fe2+ is only toxic when elevated inside cells. Interestingly, only neurons and oligodendrocytes were injured by this elevation in [Fe2+](i), whereas astrocytes were unaffected, although [Fe2+](i) was elevated to the same degree in each cell type. These studies provide a novel approach for detecting [Fe2+](i) in a manner sensitive to the redox state of the metal. These studies also provide a model system for the study of the toxic consequences of elevated [Fe2+](i) in neural cells.

Koszyca B, Manavis J, Cornish RJ, Blumbergs PC. 2002. Patterns of immunocytochemical staining for ferritin and transferrin in the human spinal cord following traumatic injury. Journal of Clinical Neuroscience 9(3): 298-301.
Abstract: Normally Fe2+ is strictly controlled within the central nervous system (CNS) because of its potential to react with oxygen and form free radicals.(1,2) Traumatic spinal cord injury (TSCI) leads to cell damage and haemorrhage, both of which may increase the pool of free iron.(3) The aim of this study was to examine the response to TSCI of the iron storage protein ferritin (Ft) and the iron transport protein transferrin (Tf). The study found a significant increase in Ft positive cells compared to controls and a significant correlation between the number of Ft positive cells and the severity of injury. Significantly fewer Tf positive cells were seen in the trauma cases compared to the control and there was no relation with the severity of injury. These observations suggest a disturbance in normal iron metabolism within the spinal cord following injury, with possible implications for free radical mediated secondary damage. (C) 2002 Published by Elsevier Science Ltd.

Knott HM, Baoutina A, Davies MJ, Dean RT. 2002. Comparative time-courses of copper-ion-mediated protein and lipid oxidation in low-density lipoprotein. Arch Biochem Biophys 400(2):223-232.
Abstract: Free radicals damage both lipids and proteins and evidence has accumulated for the presence of both oxidised lipids and proteins in aged tissue samples as well as those from a variety of pathologies including atherosclerosis, diabetes, and Parkinson's disease. Oxidation of the protein and lipid moieties of low-density lipoprotein is of particular interest due to its potential role in the unregulated uptake of lipids and cholesterol by macrophages. this may contribute to the initial stage of foam cell formation in atherosclerosis. In the study reported here, we examined the comparative time-courses of lipid and protein oxidation during copper-ion-mediated oxidation of low-density lipoprotein. We show that there is an early, lipid-mediated loss of 40-50% of the Trp residues of the apoB100 protein. There is no comparable loss over an identical period during the copper-ion-mediated oxidation of lipid-free BSA. Concomitant with Trp loss, the antioxidant alpha-tocopherol is consumed with subsequent extensive lipid peroxidation. Further changes to the protein. including the copper-ion-dependent 3.5-fold increase in 3,4-dihydroxyphenylalanine and the copper-ion-independent 3-5-fold increase in o-tyrosine, oxidation products of Tyr and Phe, respectively, only occur after maximal lipid peroxidation. Long incubation periods result in depletion of 3,4-dihydroxyphenylalanine, presumably reflecting further oxidative changes. Overall, copper-ion-mediated oxidation of LDL appears to proceed initially by lipid radical-dependent processes, even though some of the earliest detectable changes occur on the apoB100 protein. This is followed by extensive lipid peroxidation and subsequent additional oxidation of aromatic residues on apoB100, though it is not yet clear whether this late protein oxidation is

lipid-dependent or occurs as a result of direct radical attack. (C) 2002 Elsevier Science (USA). All rights reserved.

Kitazawa M, Wagner JR, Kirby ML, Anantharam V, Kanthasamy AG. 2002. Oxidative stress and mitochondrial-mediated apoptosis in dopaminergic cells exposed to methylcyclopentadienyl manganese tricarbonyl. J Pharmacol Exp Ther 302(1):26-35.
Abstract: Methylcyclopentadienyl manganese tricarbonyl (MMT), an organic manganese-containing gasoline additive, was investigated to determine whether MMT potentially causes dopaminergic neurotoxic effects. MMT is acutely cytotoxic and dopamine-producing cells (PC-12) seemed to be more susceptible to cytotoxic effects than nondopaminergic cells (striatal gamma-aminobutyric acidergic and cerebellar granule cells). MMT also potently depleted dopamine apparently by cytoplasmic vesicular release to the cytosol, a neurochemical change resembling other dopaminergic neurotoxicants. Generation of reactive oxygen species (ROS), an early effect in toxicant-induced apoptosis, occurred within 15 min of MMT exposure. MMT caused a loss of mitochondrial transmembrane potential (DeltaPsim), a likely source of ROS generation. The ROS signal further activated caspase-3, an important effector caspase, which could be inhibited by antioxidants (Trolox or N-acetyl cysteine). Predepletion of dopamine by using alpha-methyl-p-tyrosine (tyrosine hydroxylase inhibitor) treatment partially prevented caspase-3 activation, denoting a significant dopamine and/or dopamine by-product contribution to initiation of apoptosis. Genomic DNA fragmentation, a terminal hallmark of apoptosis, was induced concentration dependently by MMT but completely prevented by pretreatment with Trolox, deprenyl (monoamine oxidase-B inhibitor), and alpha-methyl-p-tyrosine. A final set of critical experiments was performed to verify the pharmacological studies using a stable Bcl-2-overexpressing PC-12 cell line. Bcl-2-overexpressing cells were significantly refractory to MMT-induced ROS generation, caspase-3 activation, and loss of DeltaPsim and were completely resistant to MMT-induced DNA fragmentation. Taken together, the results presented herein demonstrate that oxidative stress plays an important role in mitochondrial-mediated apoptotic cell death in cultured dopamine-producing cells after exposure to MMT.

Kim Y, Kim JM, Kim JW, Yoo CI, Lee CR, Lee JH, Kim HK, Yang SO, Chung HK, Lee DS, Jeon B. 2002. Dopamine transporter density is decreased in parkinsonian patients with a history of manganese exposure: What does it mean? Mov Disord 17(3):568-575.
Abstract: Manganese (Mn) exposure can cause parkinsonism. Pathological changes Occur mostly in the pallidum and striatum. Two patients with a long history of occupational Mn exposure presented with Mn-induced parkinsonism. In I patient, magnetic resonance imaging (MRI) showed findings consistent with Mn exposure, and Mn concentration was increased in the blood and urine. However, this patient's clinical features were typical of idiopathic Parkinson disease (PD). Previous pathological and positron emission tomography Studies indicate that striatal dopamine transporter density is normal in Mn-induced parkinsonism, whereas it is decreased in PD. Therefore, we performed [I-123]-(1e)-2beta-carboxymethoxy-3beta-(4([I-123]-beta-CIT) single-photon emission iodophenyl)tropane computed tomography. Severe reduction of striatal beta-CIT bindin,, was indicated, which is consistent with PD. We propose three interpretations: (1) the patients have PD, and Mn exposure is incidentals (2) Mn induces selective degeneration of presynaptic dopaminergic nerve terminals, thereby causing parkinsonism or (3) Mn exposure acts as a risk of PD in these patients. Our results and careful review of previous studies indicate that the axiom that Mn Causes parkinsonism by pallidal lesion may be over-simplified Mn exposure and parkinsonism may be more complex than previously thought. Further studies are required to elucidate the relationship between Mn and various forms of parkinsonism. (C) 2002 Movement Disorder Society.

Kim KS, Choi SY, Kwon HY, Woo MH, Kang TC, Kang JH. 2002. Aggregation of

alpha-synuclein induced by the Cu,Zn-superoxide dismutase and hydrogen peroxide system. Free Radic Biol Med 32(6):544-550.
Abstract: Alpha-synuclein is a major component of the abnormal protein aggregation in Lewy bodies of Parkinson's disease (PD) and senile plaques of Alzheimer's disease (AD). Previous studies have shown that the aggregation of alpha-synuclein was induced by copper (11) and H2O2 system. Since copper ions could be released from oxidatively damaged Cu,Zn-superoxide dismutase (SOD), we investigated the role of Cu,Zn-SOD in the aggregation of alpha-synuclein. When alpha-synuclein was incubated with both Cu,Zn-SOD and H2O2, alpha-synuclein was induced to be aggregated. This process was inhibited by radical scavengers and spin trapping agents such as 5,5'-dimethyl 1-pyrolline N-oxide and tert-butyl-alpha-phenylnitrone. Copper chelators, diethyldithiocarbamate and penicillamine, also inhibited the Cu,Zn-SOD/H2O2 system-induced alpha-synuclein aggregation. These results suggest that the aggregation of alpha-synuclein is mediated by the Cu,Zn-SOD/H2O2 system via the generation of hydroxyl radical by the free radical-generating function of the enzyme. The Cu,Zn-SOD/H2O2-induced alpha-synuclein aggregates displayed strong thioflavin-S reactivity, reminiscent of amyloid. These results suggest that the Cu,Zn-SOD/H2O2 system might be related to abnormal aggregation of alpha-synuclein, which may be involved in the pathogenesis of PD and related disorders. (C) 2002 Elsevier Science Inc.

Kim KS, Choi SY, Kwon HY, Won MH, Kang TC, Kang JH. 2002. The ceruloplasmin and hydrogen peroxide system induces alpha-synuclein aggregation in vitro. Biochimie 84(7):625-631.
Abstract: alpha-Synuclein is a key component of Lewy bodies in the brain of patients with Parkinson's disease (PD) and recent studies suggest that oxidative stress reactions might contribute to abnormal aggregation of this molecule. Since hydrogen peroxide-mediated ceruloplasmin (CP) modification can induce the formation of free radicals and release of copper ions, we investigated the role of CP in the aggregation of alpha-synuclein. When alpha-synuclein was incubated with both CP and H2O2, alpha-synuclein concomitantly was induced to be aggregated. Thioflavin-S staining of alpha-synuclein aggregates showed that they displayed characteristic fibrillar structures. Hydroxyl radical scavengers and spin-trapping agent such as 5,5'-dimethyl 1-pyrolline N-oxide and tert-butyl-alpha-phenylnitrone significantly inhibited the aggregation of alpha-synuclein. Copper chelator, penicillamine also inhibited the CP/H2O2 system-induced alpha-synuclein aggregation. This indicates that the aggregation of alpha-synuclein can be mediated by the CP/H2O2 system via the generation of hydroxyl radical. The CP/H2O2 system-induced alpha-synuclein aggregation resulted in the generation of protein carbonyl derivatives. Antioxidant molecules, carnosine, homocarnosine and anserine significantly inhibited the CP/H2O2 System-induced aggregation of alpha-synuclein. These results suggest that the CP/H2O2 system may be related to abnormal aggregation of (x-synuclein which may be involved in the pathogenesis of PD and related disorders. (C) 2002 Societe francaise de biochimie et biologic moleculaire/Editions scientifiques et medicales Elsevier SAS. All rights reserved.

Kim JR, Kwon KS, Yoon HW, Lee SR, Rhee SG. 2002. Oxidation of proteinaceous cysteine residues by dopamine-derived H2O2 in PC12 cells. Arch Biochem Biophys 397(2):414-423.
Abstract: Cellular metabolism of dopamine (DA) generates H2O2, which is further reduced to hydroxyl radicals in the presence of iron. Cellular damage inflicted by DA-derived hydroxyl radicals is thought to contribute to Parkinson's disease. We have previously developed procedures for detecting proteins that contain H2O2-sensitive cysteine (or selenocysteine) residues. Using these procedures, we identified ERP72 and ERP60, two members of the protein disulfide isomerase family, creatine kinase, glyceraldehyde-3-phosphate dehydrogenase, phospholipase C-gamma1, and thioredoxin reductase as the targets of DA-derived H2O2. Experiments with purified enzymes identified the essential Cys residues of creatine kinase and glyceraldehyde-3-phosphate dehydrogenase, that are

specifically oxidized by H2O2. Although the identified proteins represent only a fraction of the targets of DA-derived H2O2, functional impairment of these proteins has previously been associated with cell death. The oxidation of proteins that contain reactive Cys residues by DA-derived H2O2 is therefore proposed both to be largely responsible for DA-induced apoptosis in neuronal cells and to play an important role in the pathogenesis of Parkinson's disease. (C) 2002 Elsevier Science.

Kaur D, Andersen JK. 2002. Ironing out Parkinson's disease: is therapeutic treatment with iron chelators a real possibility? Aging Cell 1(1):17-21.
Abstract: Levels of iron are increased in the brains of Parkinson's disease (PD) patients compared to age-matched controls. This has been postulated to contribute to progression of the disease via several mechanisms including exacerbation of oxidative stress, initiation of inflammatory responses and triggering of Lewy body formation. In this minireview, we examine the putative role of iron in PD and its pharmacological chelation as a prospective therapeutic for the disease.

Karoline V, Gilbert R. 2002. Influence of 7,8 dihydroneopterin and hyperoxia on neurite growth and tyrosine hydroxylase activity of PC 12 cells. Pteridines 13(3):94-99.
Abstract: The influence of dihydroneopterin on tyrosine hydroxylase activity in PC 12 cells was investigated under normoxic and hyperoxic conditions, and with or without iron ions in the incubation medium, Low dihydroneopterin concentrations as well as hyperoxia increase tyrosine hydroxylase activity, Due to a too vigorous radical formation, high dihydroneopterin concentrations lead to a decrease of tyrosine hydroxylation in normoxic state and even more strongly in hyperoxic state. Similar depression of tyrosine hydroxylase activity was seen after addition of iron ions, which can form hydroxyl radicals by a Fenton type reaction. As higher iron concentrations in combination with dihydroneopterin do not suppress tyrosine hydroxylase activity completely, we conclude that dihydroneopterin and iron ions react with each other resulting in neutralisation of their effects. In conclusion, 7,8 dihydroneopterin can modulate tyrosine hydroxylase activity and its effects are dependent on concentration of oxygen as well as presence or absence of iron ions.

Karlsson J, Emgard M, Brundin P. 2002. Comparison between survival of lazaroid-treated embryonic nigral neurons in cell suspensions, cultures and transplants. Brain Res 955(1-2):268-280.
Abstract: Death of transplanted dopaminergic neurons is induced both during preparation of donor tissue and after intrastriatal grafting. Oxidative stress is thought to be partly responsible for this cell death. In the present study we compared the effects of three lipid peroxidation inhibitors, the lazaroids Tirilazad mesylate, U-83836E and U-101033, on survival of embryonic mesencephalic neurons in different paradigms. The lazaroids were equally potent in preventing serum deprivation-induced death of cultured dopaminergic neurons. In a second set of experiments, mesencephalic suspensions were pretreated with lazaroids and cell survival was analyzed immediately after dissociation, after 2 or 24 h in culture or after intrastriatal transplantation. Lazaroid pretreatment failed to protect mesencephalic neurons in the in vitro paradigms and U-101033E did not protect grafted dopaminergic neurons in contrast to the neuroprotective effects previously reported for U-83836E and Tirilazad. Pretreatment with the iron chelator deferoxamine mesylate did not protect cultured or grafted dopaminergic neurons, nor did it improve neuronal survival in the serum deprivation model. U-83836E and U-101033E, but not Tirilazad, prevented cell death induced by the pro-oxidant tert-butyl hydroperoxide in suspensions. In a final experiment, we found that systemic treatment of the graft recipient rat with Tirilazad mesylate (before and during the first 3 days after grafting) improved survival of transplanted dopaminergic neurons to 180% of control values. Our results show that systemic treatment with a lipid peroxidation inhibitor for 3 days can promote graft survival, but also highlights the poor correlation between neuroprotective effect of pharmacological compounds in vitro and in grafts. (C) 2002

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Jurva U, Wikstrom HV, Bruins AP. 2002. Electrochemically assisted Fenton reaction: reaction of hydroxyl radicals with xenobiotics followed by on-line analysis with high-performance liquid chromatography/tandem mass spectrometry. Rapid Commun Mass Spectrom 16(20):1934-1940.
Abstract: Oxygen radicals are generated in vivo by various processes, often as toxic intermediates in different metabolic transformations, and have been shown to play an important role for a large number of diseases. In this article we introduce an electrochemical flow-through system that allows generation of hydroxyl radicals for reaction with xenobiotics and subsequent detection of the oxidation products on-line with high-performance liquid chromatography/tandem mass spectrometry (HPLC/NIS/MS). The system is based on the Fenton reaction and is predominantly aimed at the generation of hydroxyl radicals; however, by minor variations to the system, a broad range of other radicals can be produced. Optimization of the system was performed with the radical scavenger 5,5-dimethyl-1-pyrroline-N-oxide (DMPO). Under the same physical conditions, one injection through the electrochemical cell gave a higher yield of the oxidation product N-hydroxy-5,5-dimethylpyrrolidin-2-one than what was attained after 60 min with a chemical Fenton system catalyzed by ascorbic acid. Since the iron is added as Fe3+ the initial mixture is 'inactive' until it reaches the electrochemical cell. This makes it very suitable for on-line analysis of the generated compounds, since the whole reaction mixture, including substrate, can be kept in a vial in an autosampler. The system described provides a useful tool for investigation of new radical scavengers and antioxidants. Since the hydroxyl radical adds readily to unsaturated pi-systems, the technique is also suitable for on-line generation and characterization of potential drug metabolites resulting from hydroxylation of double bonds and aromatic systems. Copyright (C) 2002 John Wiley Sons, Ltd.

Jellinger KA. 2002. Recent developments in the pathology of Parkinson's disease. Journal of Neural Transmission-Supplement (62):347-376.
Abstract: Parkinson's disease (PD) is morphologically characterized by progressive loss of neurons in the substantia nigra pars compacta (SNpc) and other subcortical nuclei associated with intracytoplasmic Lewy bodies and dystrophic (Lewy) neurites mainly in subcortical nuclei and hippocampus und, less frequently in cerebral cortex. SN cell loss is significantly related to striatal dopamine (DA) deficiency as well as to both the duration and clinical severity of disease, The two major clinical subtypes of PD show different morphologic lesion patterns: the akinetic-rigid form has more severe cell loss in the ventrolateral part of SN with negative correlation to DA loss in the posterior putamen, and motor symptoms related to overacitivty of the GABAergic "indirect" motor loop, which causes inhibition of the glutamatergic thalamocortical pathway and reduced cortical activation. The tremor-dominant type shows more severe cell loss in the medial SNpc and retrorubal field A 8, which project to the matrix of the dorsolateral striatum and ventromedial thalamus, thus causing hyperactivity of thalamornotor and cerebellar projections. These and experimental data suggesting different pathophysiological mechanisms for the major clinical subtypes of PD may have important therapeutic implications. Lewy bodies, the morphologic markers of PD, are composed of hyperphosphorylated neurofilament proteins, lipids, redox-active iron, ubiquitin, and alpha-synuclein, showing a continuous accumulation in the periphery and of ubiquitin in the central core. alpha-synuclein, is usually unfolded in alpha-helical form. By gene mutation, environmental stress or other factors it can be transformed to beta-folding which is sensible to self-aggregation in filamentous fibrils and formation of insoluble intracellular inclusions that may lead to functional disturbances and, finally, to death of involved neurons. While experimental and tissue culture studies suggest that apoptosis, a genetically determined form of programmed cell death, represents the most common pathway in neurodegeneration, DNA fragmentation, overexpression of proapoptotic proteins and activated caspase-3, the effector enzyme of the terminal apopoptic cascade, have

only extremely rarely been detected in SN of PD brains. This is in accordance with the rapid course of apoptotis and the extremely slow progression of the neurodegenerative process in PD. The biological role of Lewy bodies and other intracellular inclusions, the mechanisms of the intracellular aggregation of insoluble protein deposits, and their implication for cellular dysfunction resulting in neurodegeneration and cell demise are still unresolved. Further elucidation of the basic molecular mechanisms of cytoskeletal lesions will provide better insight into the pathogenesis of neurodegeneration in PD and related disorders.

Ikegami T, Suzuki Y, Shimizu T, Isono K, Koseki H, Shirasawa T. 2002. Model mice for tissue-specific deletion of the manganese superoxide dismutase (MnSOD) gene. Biochem Biophys Res Commun 296(3):729-736.
Abstract: Manganese superoxide dismutase (MnSOD) is the enzyme that converts toxic O-2(-) to H2O2 in mitochondria. Previous reports showed that a deficiency of MnSOD in mice was neonatal lethal. Therefore, a model mouse was not available for the analysis of the pathological role of O-2(-) injuries in adult tissues. To explore an adult-type model mouse, we designed tissue-specific MnSOD conditional knockout mice using a Cre-loxp system. First, we crossbred MnSOD flox mice with transgenic mice expressing Cre recombinase under the control of the chicken actin promoter (CAG). We confirmed that CAG MnSOD knockout mice were completely deficient in MnSOD and died as neonates, validating the use of the Cre-loxp system. Next, we generated liver-specific MnSOD-deficient mice by crossbreeding with Alb-Cre transgenic mice. MnSOD activity and protein were both significantly downregulated. in the liver of liver-specific MnSOD knockout mice. However, no obvious morphological abnormality was observed in the liver when biochemical alterations such as lipid peroxidation were not detectable, suggesting a redundant or less important physiological role for MnSOD in the liver than previously thought. In the present study, we successfully generated tissue-specific MnSOD conditional knockout mice that would provide a useful tool for the analysis of various age-associated diseases such as diabetes mellitus, Parkinson's disease, stroke, and heart disease, when crossbred with tissue-specific transgenic Cre mice. (C) 2002 Elsevier Science (USA). All rights reserved.

Ide-Ektessabi A, Fujisawa S, Yoshida S. 2002. Chemical state imaging of iron in nerve cells from a patient with Parkinsonism-dementia complex. Journal of Applied Physics 91(3):1613-1617.
Abstract: X-ray fluorescence spectroscopy and Fe K-edge x-ray absorption near-edge structure spectroscopy were performed on postmortem human tissues containing nerve cells in order to investigate distributions and chemical states of iron. Specimens used in this study were obtained from the substantia nigra of a patient with Parkinsonism-dementia complex (PDC) of Guam and a control subject. Iron concentration was observed in the neuromelanin granules and in one of the glial cells surrounding the neuromelanin granules of the PDC patient. Iron was also detected in melanized neurons of the control subject. Chemical state imaging which separates Fe2+ and Fe3+ in iron compounds showed that the glial cell of the PDC patient has a higher concentration of Fe3+. Iron contained in the neuromelanin granules of the PDC patient was mixed states of Fe2+ and Fe3+. (C) 2002 American Institute of Physics.

Hynes MJ, Coinceanainn MO. 2002. Investigation of the release of iron from ferritin by naturally occurring antioxidants. J Inorg Biochem 90(1-2):18-21.
Abstract: Ferritin is the main intracellular iron storage protein. The release of iron from ferritin in the presence of a number of phenolic based compounds of nutritional significance was studied at physiological pH. The release of iron was measured by monitoring the formation of the iron(II)-ferrozine complex. The kinetics of this process were studied in Hepes buffer (pH 7.00), at 37 degreesC, The order of ability to remove iron from ferritin is epigallocatechin>gallic acid methyl ester-sinapic acid>ferulic acid. The presence of the oxyradical scavenger urea resulted in a slight inhibition in the release of iron from ferritin by both gallic acid methyl ester and epigallocatechin. The ability of each reagent to release iron is

interpreted on the basis of their ability to (a) reduce the bound iron and (b) complex the iron with the oxidised form of the phenol, thus mobilising it from the protein. These studies indicate that some phenolic based compounds that have been epidemiologically associated with a negative effect on iron absorption in man, can individually mobilise and release iron from ferritin under suitable conditions. (C) 2002 Elsevier Science Inc. All rights reserved.

Hussain S, Ali SF. 2002. Zinc potentiates 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine induced dopamine depletion in caudate nucleus of mice brain. Neurosci Lett 335(1):25-28.
Abstract: Present study describes the effect of zinc (Zn) on 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) induced dopamine depletion in mice brain. MPTP is a known neurotoxicant primarily causing marked depletion of dopamine (DA) levels in nigrostriatal dopaminergic system. Adult Male C57-mice were intraperitonially injected with 25 mg/kg MPTP in the presence or absence of zinc acetate. Twenty-four hours after treatment animals were sacrificed and DA levels were determined by high performance liquid chromatography in caudate nucleus of control and treated mice. The results showed that there was a marked depletion of DA in MPTP treated mice, whereas no change was observed in DA levels in mice treated with Zn when compare to controls. Interestingly, mice receiving MPTP in conjunction with Zn showed significantly lower DA levels in brain when compare to animals receiving MPTP alone. In summary the data suggest that Zn treatment potentiates depletion of dopamine in MPTP treated mice. (C) 2002 Elsevier Science Ireland Ltd. All rights reserved.

Hirata Y. 2002. Manganese-induced apoptosis in PC12 cells. Neurotoxicol Teratol 24(5):639-653.
Abstract: Manganese has been known to induce neurological disorders similar to parkinsonisms for a long time. Dopamine deficiency has been demonstrated in Parkinson's disease and in chronic manganese poisoning, suggesting that the mechanisms underlying the neurotoxic effects of the metal ion are related to dysfunction of the extrapyramidal system. However, the details of the mechanisms have yet to be elucidated. In an effort to learn more about the toxicity of manganese, we have employed an in vitro model that uses the PC12 catecholaminergic cell line. In this model, manganese induces apoptosis in PC12 cells. In this paper, experiments conducted with this model, the cellular biochemical changes, and the mechanism of the cell death are reviewed. (C) 2002 Elsevier Science Inc. All rights reserved.

Hernandez EH, Valentini MC, Discalzi G. 2002. T1-weighted hyperintensity in basal ganglia at brain magnetic resonance imaging: Are different pathologies sharing a common mechanism? Neurotoxicology 23(6):669-674.
Abstract: Basal ganglia bilateral symmetric hyperintensity in T1-weighted sequences at magnetic resonance imaging (MRI) is recognized to be due to the presence of manganese deposits. This abnormal finding has been reported in occupational exposures, liver cirrhosis and total parenteral nutrition with unbalanced solutions. However, the same imaging is often observed "by chance" in brain MRIs of patients not belonging to these groups. In order to better understand which are the clinical conditions coexisting with such findings, we decided to study systematically patients which showed this kind of imaging, focusing on their manganese and iron status, as it is known that these two metals have similar properties and that iron-deficiency can competitively increase manganese absorption. The 20 patients studied underwent clinical evaluation and the following laboratory tests: whole blood iron and manganese, hemoglobin, plasma iron, transferrin and ferritin. The neuroradiologic evaluation was integrated by pallidal index calculation, in order to provide a semi-quantitative esteem of the hyperintensity. The patients could be classified into four subgroups: Parkinsonism, anemia, cirrhosis, central nervous system tumors. In 18 out of 20 cases, we found abnormalities in iron and/or manganese-related values. Particularly, iron-deficiency seems to be frequent among patients showing brain MRI abnormalities compatible with manganese deposits in

basal ganglia. This observation suggests that iron-deficiency could be an important risk-factor for manganese-induced neurotoxicity and should, therefore, be accurately considered and treated. (C) 2002 Elsevier Science Inc. All rights reserved.

Hermann W, Eggers B, Barthel H, Clark D, Villmann T, Hesse S, Grahmann F, Kuhn HJ, Sabri O, Wagner A. 2002. Correlation between automated writing movements and striatal dopaminergic innervation in patients with Wilson's disease. J Neurol 249(8):1082-1087.
Abstract: Handwriting defects are an early sign of motor impairment in patients with Wilson's disease. The basal ganglia being the primary site of copper accumulation in the brain suggests a correlation with lesions in the nigrostiatal dopaminergic system. We have analysed and correlated striatal dopaminergic innervation using [I-123] beta-CIT-SPECT and automated handwriting movements in 37 patients with Wilson's disease. There was a significant correlation of putaminal dopaminergic innervation with fine motor ability ($p < 0,05$ for NIV [number of inversion in velocity], NIA [number of inversion in acceleration], frequency). These data suggest that loss of dorsolateral striatal dopaminergic innervation has a pathophysiological function for decreased automated motor control in Wilson's disease. Furthermore analysis of automated handwriting movements could be useful for therapy monitoring and evaluation of striatal dopaminergic innervation.

He Y, Lee T, Leong SK, Le WD. 2002. The onset of dopaminergic cell death in the substantia nigra of 1-methyl-4-pheyl-1,2,3,6-tetrahydropyridine (MPTP)-induced parkinsonian model of monkeys precedes the elevation of iron concentration. Neurology 58(7):A373-A374.

Gwiazda RH, Lee D, Sheridan J, Smith DR. 2002. Low cumulative manganese exposure affects striatal GABA but not dopamine. Neurotoxicology 23(1): 69-76.
Abstract: The introduction of the anti-knock methylcyclopentadienyl manganese (Mn) tricarbonyl (MMT) in gasoline has raised concerns about the potential for manganese neurotoxicity. Because subpopulations such as the elderly in the early stages of neurodegenerative disease may be at increased risk for manganese toxicity, a pre-Parkinsonism rat model was used to evaluate whether sub-chronic manganese exposure can aggravate the neurochemical and behavioral dysfunctions characteristic of Parkinsonism. Sub-threshold levels of dopamine depletion of 3.5, 53 and 68% were generated via intrastriatal unilateral 6-hydroxydopamine (6-OHDA) doses. A sub-chronic dosing regimen of low cumulative manganese exposure (4.8 mg Mn/kg body weight, 3 i.p. injections per week x 5 weeks) was started 4 weeks after 6-OHDA treatments. Neurochemical and neuromotor (functional observational battery (FOB)) measures were evaluated. Manganese produced significant ($P < 0.05$) reductions of 30-60% in motor function. This effect was exacerbated in the presence of a pre-Parkinsonism condition [Neurotox. Teratol. 22 (2000) 851]. Manganese did not affect striatal dopamine, but resulted in significant increases in striatal γ-aminobutyric acid (GABA) of 16 and 22% ($P < 0.01$) in both striati and a borderline non-significant 4% increase in frontal cortex ($P = 0.076$). Manganese treatment produced increased aspartate ($P < 0.01$) in the manganese and 6-OHDA treated striatum. In light of previous studies predominantly showing dopamine depletion with elevated manganese exposures, the significant effects of manganese on striatal GABA but not on striatal dopamine at the low cumulative exposure administered here suggest a progression in manganese toxicity with increasing cumulative dose, whereby GABA levels are adversely affected before striatal dopamine levels. Because these neurochemical disruptions were accompanied by motor dysfunction that was exacerbated in the presence of a pre-Parkinsonism condition, an increased environmental burden of manganese may have deleterious effects on populations with sub-threshold neurodegeneration in the basal ganglia (e.g. pre-Parkinsonism). (C) 2002 Elsevier Science Inc. All rights reserved.

Green H, Ross G, Peacock J, Owen R, Yarnold J, Houlston R. 2002. Variation in the manganese superoxide dismutase gene (SOD2) is not a major cause of radiotherapy complications in breast cancer patients. Radiother Oncol 63 (2):213-216.
Abstract: Background and purpose: Small proportions of patients receiving radiotherapy develop marked long-term radiation damage. It is thought that this is due, at least in part, to intrinsic differences in cellular radiosensitivity, but the underlying mechanism is unknown. Reactive oxygen species are involved in cellular radiation damage, hence inter-individual differences in free radical detoxification may be related to radiosensitivity. Within mitochondria manganese superoxide dismutase (MnSOD) provides a major defence against oxidative damage by reactive oxygen species. MnSOD has been linked to expression of malignant phenotype and apoptosis and polymorphic variation in the gene, SOD2 to risk of breast cancer. Materials and methods: Forty-one breast cancer patients developing marked changes in breast appearance after radiotherapy and 39 patients who showed no clinically detectable reaction after radiotherapy were analyzed for germline sequence variation in SOD2. Results: The Ala-9Val polymorphism was detected, but no other sequence variants were detected in SOD2. Both alleles of the Ala-9Val polymorphism were equally distributed between the two patient groups. Conclusions: Sequence variation in SOD2 is not the major cause of radiotherapy complications in women with breast cancer. (C) 2002 Elsevier Science Ireland Ltd. All rights reserved.

Graumann R, Paris I, Martinez-Alvarado P, Rumanque P, Perez-Pastene C, Cardenas SP, Marin P, Diaz-Grez F, Caviedes R, Caviedes P, Segura-Aguilar J. 2002. Oxidation of dopamine to aminochrome as a mechanism for neurodegeneration of dopaminergic systems in Parkinson's disease. Possible neuroprotective role of DT-diaphorase. Pol J Pharmacol 54(6): 573-579.
Abstract: Although it is generally accepted that free radicals are involved in the neurodegenerative process occurring in the dopaminergic neurons of the nigro-striatal system in Parkinson's disease, the exact mechanism of neurodegeneration in vivo is still unknown. We propose that the degeneration of dopaminergic nigrostriatal system in this condition may depend on: (a) existence of free dopamine which oxidizes to aminochrome as a consequence of. (i) overproduction of dopamine; (h) inhibition and/or low expression of synaptic vesicle catecholamine transporter; (iii) inhibition or low expression of monoamine oxidases; (b) one-electron reduction of aminochrome to leukoaminochrome o-semiquinone radical, which induces neurotoxicity, due to inhibition of DT-diaphomse or the existence of a polymorphism with a point mutation (C --> T) in the cDNA 609 expressing an inactive DT-diaphorase. We suggest that DT-diaphorase plays a neuroprotective role in dopaminergic neurons, which is supported by the following observations: (i) Cu-toxicity is dependent on DT-diapborase inhibition with dicoumarol in RCSN-3 cells derived from the rat substantia, nigra; (ii) the cytotoxic effect of monoamine oxidase-A inhibitor amiflamine in RCSN-3 cells is increased by 2.4-fold ($p < 0.001$) in the presence of the inhibitor of DT-diaphorase, dicoumarol; (iii) concomitant intracerebral administration of manganese (Mn3+) together with the DT-diaphorase inhibitor dicoumarol into the left medial forebrain bundle produced a behavioral pattern characterized by contralateral rotational behavior when the rats were stimulated with apomorphine, in a manner similar to that observed in animals injected unilaterally with 6-hydroxydopamine; (iv) incubation of RCSN-3 cells with salsolinol in the presence of DT-diaphorase inhibitor significantly decreased cell survival by 2.5-fold ($p < 0.001$).

Golts N, Snyder H, Frasier M, Theisler C, Choi P, Wolozin B. 2002. Magnesium inhibits spontaneous and iron-induced aggregation of alpha-synuclein. J Biol Chem 277(18):16116-16123.
Abstract: Multiple studies implicate metals in the pathophysiology of neurodegenerative diseases. Disturbances in brain iron metabolism are linked with synucleinopathies. For example, in Parkinson's disease, iron levels are increased and magnesium levels are reduced in the brains of

patients. To understand how changes in iron and magnesium might affect the pathophysiology of Parkinson's disease, we investigated binding of iron to alpha-synuclein, which accumulates in Lewy bodies. Using fluorescence of the four tyrosines in alpha-synuclein as indicators of metal-related conformational changes in alpha-synuclein, we show that iron and magnesium both interact with alpha-synuclein. alpha-Synuclein exhibits fluorescence peaks at 310 and 375 nm. Iron lowers both fluorescence peaks, while magnesium increases the fluorescence peak only at 375 nm, which suggests that magnesium affects the conformation of alpha-synuclein differently than iron. Consistent with this hypothesis, we also observe that magnesium inhibits alpha-synuclein aggregation, measured by immunoblot, cellulose acetate filtration, or thioflavine-T fluorescence. In each of these studies, iron increases alpha-synuclein aggregation, while magnesium at concentrations >0.75 mM inhibits the aggregation of alpha-synuclein induced either spontaneously or by incubation with iron. These data suggest that the conformation of alpha-synuclein can be modulated by metals, with iron promoting aggregation and magnesium inhibiting aggregation.

Gerber GB, Leonard A, Hantson P. 2002. Carcinogenicity, mutagenicity and teratogenicity of manganese compounds. Crit Rev Oncol Hematol 42(1): 25-34.
Abstract: Manganese, an essential trace element, is one of the most used metals in the industry. Recently, several new manganese compounds have been introduced as fungicide, as antiknock agent in petrol and as contrasting agent in nuclear magnetic resonance tomography. Manganese displays a somewhat unique behaviour with regard to its toxicity. It is relatively non-toxic to the adult organism except to the brain where it causes Parkinson-like symptoms when inhaled even at moderate amounts over longer periods of time. Relatively high doses of manganese affect DNA replication and repair in bacteria and causes mutations in microorganism and mammalian cells although the Ames test does not appear to be particularly responsive to manganese. In mammalian cells, manganese causes DNA damage and chromosome aberrations. Information on organic manganese derivatives is still insufficient. Large amounts of manganese affect fertility in mammals and are toxic to the embryo and foetus. The fungicide MANEB and the contrasting agent MnDPDP also can be embryotoxic, but the latter only at doses much higher than those clinically employed, Information on the anti-knock agent MMT is inadequate. On the other hand, manganese deficiency can also affect fertility and be teratogenic. Information on cancer due to manganese is scanty but the results available do not indicate that inorganic manganese is carcinogenic. More information is desirable with regard to the organic manganese derivatives. It may surprise that an agent that causes mutations is not also carcinogenic. The experience with manganese shows that conclusions with regard to carcinogenicity of an agent based on the observation of mutations are subject to uncertainties. Altogether, it appears that, because of the very high doses at which positive effects hake been found, manganese would not represent a significant carcinogenic risk to the population and workers. Care must, however, be exercised with respect to central-nervous symptoms after chronic exposure and with respect to effects on the embryo. Pregnant women should not be exposed to manganese at the work place. (C) 2002 Elsevier Science Ireland Ltd. All rights reserved.

Gearing M, Juncos JL, Procaccio V, Gutekunst CA, Marino-Rodriguez EM, Gyure KA, Ono S, Santoianni R, Krawiecki NS, Wallace DC, Wainer BH. 2002. Aggregation of actin and cofilin in identical twins with juvenile-onset dystonia. Ann Neurol 52(4):465-476.
Abstract: The neuropathology of the primary dystonias is not well understood. We examined brains from identical twins with DYT1-negative, dopa-unresponsive dystonia. The twins exhibited mild developmental delays until age 12 years when they began developing rapidly progressive generalized dystonia. Genetic, metabolic, and imaging studies ruled out known causes of dystonia. Cognition was subnormal but stable until the last

few years. Death occurred at ages 21 and 22 years. The brains were macroscopically unremarkable. Microscopic examination showed unusual glial fibrillary acidic protein-immunoreactive astrocytes in multiple regions and iron accumulation in pallidal and nigral neurons. However, the most striking findings were 1) eosinophilic, rod-like cytoplasmic inclusions in neocortical and thalamic neurons that were actin depolymerizing factor/ cofilin-immunoreactive but only rarely actin-positive; and 2) abundant eosinophilic spherical structures in the striatum that were strongly actin- and actin depolymerizing factor/cofilin-positive. Electron microscopy suggested that these structures represent degenerating neurons and processes; the accumulating filaments had the same dimensions as actin microfilaments. To our knowledge, aggregation of actin has not been reported previously as the predominant feature in any neurodegenerative disease. Thus, our findings may shed light on a novel neuropathological change associated with dystonia that may represent a new degenerative mechanism involving actin, a ubiquitous constituent of the cytoskeletal system.

Galazka-Friedman J, Bauminger ER, Friedman A . 2002. Iron in Parkinson's disease revisited. Hyperfine Interactions 141(1-4):267-271.
Abstract: Mossbauer studies of fresh frozen samples taken at autopsy from different parts of the human brain (globus pallidus (GP), substantia nigra (NS), and hippocamp (Hip)) showed a relatively high concentration of iron in these structures. Mossbauer data, biochemical results and transmission electron micrographs lead to the conclusion that in all above-mentioned structures iron is located mainly within ferritin. However, the Mossbauer doublets obtained from most brain samples at 90 K are slightly asymmetric. This asymmetry could be caused by the presence of a small amount of non-ferritin-like iron. Measurements at 4.1 K showed besides the six-line spectra characteristic for ferritin-like iron, an additional doublet with Mossbauer parameters different from ferritin. We found a slightly higher asymmetry and intensity of the 4.1 K doublet in Mossbauer spectra of Parkinsonian SN than in control SN. As Parkinson's disease is a progressive degeneration of nervous cells in SN and iron may be involved in this degeneration process, this may suggest that the factors evoking these phenomena are related to the pathogenesis of Parkinson's disease.

Fukuyama R, Nakayama A, Nakase T, Toba H, Mukainaka T, Sakaguchi H, Saiwaki T, Wada M, Sakurai H, Fushiki S. 2002. A newly established neuronal rho-0 cell line highly susceptible to oxidative stress accumulates iron and other metals - Relevance to the origin of metal ion deposits in brains with neurodegenerative disorders. J Biol Chem 277(44): 41455-41462.
Abstract: From human neuroblastoma-derived SILA cells we have established a p-0 cell line that is deficient in both respiration and mitochondrial DNA. Lactate dehydrogenase activity, lactate production, and growth in the medium without glucose indicate that these cells shift from aerobic to anaerobic metabolism. Electron microscopic observations revealed abnormal mitochondria with unique cristae structures. Staining with MitoTracker dye showed that the mitochondrial transmembrane potential was reduced by 30-40% from the parent cell levels. These cells were markedly susceptible to H2O2 and died apparently by a necrotic mechanism, a process blocked by deferoxamine in the parent cells but not p-0 cells. Analysis by inductively coupled plasma-mass spectrometry revealed an approximately 3-fold accumulation of iron in the p-0 cells at confluence (n = 4-6, three clones, *p < 0.05). Iron and four other metals were all elevated in the cells of one of the p-0 clones and were similar to control levels in the control cybrid cells, which were replenished with normal mitochondrial DNA. Their sensitivity to H2O2 was also similar to that of the parent cells. These results indicate that a newly established neuronal related p-0 cell line is highly susceptible to active oxygen species and that these toxicity effects appear to be related to an accumulation of transition metals, which probably occurs through the respiratory impairment.

Fitsanakis VA, Amarnath V, Moore JT, Montine KS, Zhang J, Montine TJ. 2002. Catalysis of catechol oxidation by metal-dithiocarbamate complexes in pesticides. Free Radic Biol Med 33(12):1714-1723.
Abstract: Dithiocarbamate (DTC)-based pesticides have been implicated in Parkinson's disease (PD) through epidemiological links to increased risk of PD, clinical reports of parkinsonism following occupational exposure to the DTC-based pesticide maneb, and experimental studies showing dopaminergic neurodegeneration with combined exposure of rats to maneb and paraquat. We hypothesize that the manganese-ethylene-bis-dithiocarbamate (MnEBDC) complex in maneb may produce oxidative stress by catalyzing catechol oxidation. We tested this hypothesis by performing a structure-function analysis of metal-EBDC and metal-diethyldithiocarbamate (DEDC) complexes of Mn2+, Zn2+, and Cu2+ to catalyze oxidation of N-acetyldopamine (NA-DA) and 3,4-dihydroxyphenyl acetic acid (DP) in the presence and absence of N-acetylcysteine (NAC), a model of glutathione. Both Mn-DTCs retained the capacity of the parent ion to catalyze one-electron oxidation of NA-DA, but lost the ability to catalyze DP oxidation. Strikingly, while Zn2+ did not catalyze catechol oxidation, both Zn-DTCs catalyzed one-electron oxidation of NA-DA but not DP. While Cu2+ catalyzed oxidation of both catechols, Cu-DTCs were inert. Similar results were obtained with MnEBDC and dopamine or norepinephrine; however, zinc-ethylene-bis-dithiocarbamate was less efficient at catalyzing oxidation of these catechols. Our results point to the potential for manganese- and zinc-containing EBDC pesticides to promote oxidative stress in catecholaminergic regions of the brain. (C) 2002 Elsevier Science Inc.

Finkelstein Y, Vardi J. 2002. Progressive parkinsonism in a young experimental physicist following long-term exposure to methanol. Neurotoxicology 23 (4-5):521-525.
Abstract: A case is described of an experimental physicist who developed parkinsonism, apparently as delayed toxic effect of long exposure to vapors of methanol in the laboratory. Clinical and magnetic resonance imaging (MRI) supported the diagnosis, after exclusion of hereditary diseases and primary degenerative diseases. Screening for heavy metals in urine and plasma ceruloplasmin was negative. This case illustrates the neurotoxic delayed effect of long-term exposure to methanol with no episodes of acute intoxication. The setting of a research laboratory with prolonged exposure to mixed single crystals and inhalation of methanol vapors may exist in other academic and hi-tech environments, and pose the risk of similar delayed toxic influences. (C) 2002 Elsevier Science Inc. All rights reserved.

Figueiredo-Pereira ME, Li ZM, Jansen M, Rockwell P. 2002. N-acetylcysteine and celecoxib lessen cadmium cytotoxicity which is associated with cyclooxygenase-2 up-regulation in mouse neuronal cells. J Biol Chem 277 (28):25283-25289.
Abstract: In many neurodegenerative disorders, aggregates of ubiquitinated proteins are detected in neuronal inclusions, but their role in neurodegeneration remains to be defined. To identify intracellular mechanisms associated with the appearance of ubiquitin-protein aggregates, mouse neuronal HT4 cells were treated with cadmium. This heavy metal is a potent cell poison that mediates oxidative stress and disrupts the ubiquitin/ proteasome pathway. In the current studies, the following intracellular events were found to be also induced by cadmium: (i) a specific rise in cyclooxygenase-2 (COX-2) gene expression but not COX-1; (ii) an increase in the extracellular levels of the proinflammatory prostaglandin E2, a product of COX-2; and (iii) production of 4-hydroxy-2-nonenal-protein adducts, which result from lipid peroxidation. In addition, cadmium treatment led to the accumulation of high molecular weight ubiquitin COX-2 conjugates and perturbed COX-2 glycosylation. The thiol-reducing antioxidant N-acetylcysteine, and, to a lesser extent, the COX-2 inhibitor celecoxib, attenuated the loss of cell viability induced by cadmium demonstrating that oxidative stress and COX-2 activation contribute to cadmium cytotoxicity. These findings establish that disruption of the

ubiquitin/proteasome pathway is not the only event triggered by cadmium. This oxidative stressor also activates COX-2 function. Both events could be triggered by formation of 4-hydroxy-2-nonenal as a result of cadmium-induced lipid peroxidation. Proinflammatory responses stimulated by oxidative stressors that mimic the cadmium effects may, therefore, be important initiators of the neurodegenerative process and exacerbate its progress.

Faucheux BA, Martin ME, Beaumont C, Hunot S, Hauw JJ, Agid Y, Hirsch EC. 2002. Lack of up-regulation of ferritin is associated with sustained iron regulatory protein-1 binding activity in the substantia nigra of patients with Parkinson's disease. J Neurochem 83(2):320-330.
Abstract: Dopaminergic neurones degenerate during Parkinson's disease and cell loss is most extensive in the subpopulation of melanized neurones located in the substantia nigra pars compacta. Iron accumulation, together with a lack of up-regulation of the iron-storing protein, ferritin, has been reported and may contribute to increased oxidative stress in this region. We investigated the binding activity of iron regulatory protein-1 (IRP1) to the iron-responsive element that precludes ferritin mRNA translation, in the substantia nigra of a group of parkinsonian patients who presented a statistically significant reduction in the number of nigral melanized-neurones and an increased iron content, together with unchanged H-ferritin and L-ferritin subunit levels as compared to matched controls. The levels of ferritin mRNAs and the binding activity of IRP1 to the iron-responsive element of ferritin mRNA did not differ significantly between the two groups. Moreover, there was no detectable contribution of the iron regulatory protein-2 (IRP2) binding activity. No change in IRP1 control of ferritin mRNA translation explains the lack of up-regulation of ferritin expression in cytoplasmic extracts of SNpc that would be normally expected with cytosolic iron accumulation. The data of this study do not favor changes in transcription and post-transcriptional regulation of ferritin expression in Parkinson's disease and suggest a 'compartmentalized' iron accumulation.

El-Agnaf OMA, Irvine GB. 2002. Aggregation and neurotoxicity of alpha-synuclein and related peptides. Biochem Soc Trans 30:559-565.
Abstract: Fibrillar deposits of alpha-synuclein occur in several I neurodegenerative diseases. Two mutant forms of alpha-synuclein have been associated with earlv-onset Parkinson's disease, and a fragment has been identified as the non-amyloid-beta peptide component of Alzheimer's disease amyloid (NAC). Upon aging, solutions of alpha-synuclein and NAC change conformation to beta-sheet, detectable by CD spectroscopy, and form oligomers that deposit as amyloid-like fibrils, detectable by electron microscopy. These aged peptides are also neurotoxic. Experiments on fragments of NAC have enabled the region of NAC responsible for its aggregation and toxicity to be identified. NAC(8-18) is the smallest fragment that aggregates, as indicated by the concentration of peptide remaining in solution after 3 days, and forms fibrils, as determined by electron microscopy. Fragments NAC(8-18) and NAC(8-16) are toxic, whereas NAC(12-18), NAC(9-16) and NAC(8-15) are not. Hence residues 8-16 of NAC comprise the region crucial for toxicity. Toxicity induced by alpha-synuclein, NAC and NAC(1-18) oligomers occurs via an apoptotic mechanism, possibly initiated by oxidative damage, since these peptides liberate hydroxyl radicals in the presence of iron. Molecules with anti-aggregational and/or antioxidant properties may therefore be potential therapeutic agents.

Ebadi M, Shavali S. 2002. Salsolinol, a neurotoxin causing Parkinsonism decreases the expression of metallothionein and BCL-2 in human dopaminergi. J Neurochem 81:78.

Ebadi M, Sharma SK. 2002. Mitochondrial alpha-synuclein-metallothionein interaction in Parkinson's disease. FASEB J 16(5):A950.

Ebadi M, Sharma S, Shavali S, El Refaey H. 2002. Neuroprotective actions of

selegiline. J Neurosci Res 67(3):285-289.
Abstract: Selegiline, a selective inhibitor of monoamine oxidase-B (MAO-B), was one of the first adjunct therapies in clinical neurology. A retrospective analysis of data from patients with Parkinson's disease found a significant increase in survival in those treated with selegiline plus L-dopa compared with L-dopa alone. The mechanism of action of selegiline is complex and cannot be explained solely by its MAO-B inhibitory action. Pretreatment with selegiline can protect neurons against a variety of neurotoxins, such as 1-methyl-4-phenyl-1,2,3,6 tetrahydropyridine (MPTP), 6-hydroxydopamine, N-(2-chloroethyl)-N-ethyl-2-bromobenzylamine (DSP-4), methyl-beta-acetoxyethyl-2-chloroethylamine (AF64A), and 5,6-dihydroxyserotonin, which damage dopaminergic, adrenergic, cholinergic, and sertoninergic neurons, respectively. Selegiline produces an amphetamine-like effect, enhances the release of dopamine, and blocks the reuptake of dopamine. It stimulates gene expression of L-aromatic amino acid decarboxylase, increases striatal phenylethylamine levels, and activates dopamine receptors. Selegiline reduces the production of oxidative radicals, up-regulates superoxide dismutase and catalase, and suppresses nonenzymatic and iron-catalyzed autooxidation of dopamine. Selegiline compensates for loss of target-derived trophic support, delays apoptosis in serum-deprived cells, and blocks apoptosis-related fall in the mitochondrial membrane potential. Most of the aforementioned properties occur independently of selegiline's efficacy to inhibit MAO-B. (C) 2002 Wiley-Liss, Inc.

Du S, Mclaughlin B, Pal S, Aizenman E. 2002. In vitro neurotoxicity of methylisothiazolinone, a commonly used industrial and household biocide, proceeds via a zinc and extracellular signal-regulated kinase mitogen-activated protein kinase-dependent pathway. J Neurosci 22(17): 7408-7416.
Abstract: Neurodegenerative disorders in humans may be triggered or exacerbated by exposure to occupational or environmental agents. Here, we show that a brief exposure to methylisothiazolinone, a widely used industrial and household biocide, is highly toxic to cultured neurons but not to glia. We also show that the toxic actions of this biocide are zinc dependent and require the activation of p44/42 extracellular signal-regulated kinase (ERK) via a 12-lipoxygenase-mediated pathway. The cell death process also involves activation of NADPH oxidase, generation of reactive oxygen species, DNA damage, and overactivation of poly(ADP-ribose) polymerase, all occurring downstream from ERK phosphorylation. The toxic effects of methylisothiazolinone and related biocides on neurons have not been reported previously. Because of their widespread use, the neurotoxic consequences of both acute and chronic human exposure to these toxins need to be evaluated.

Dror N, Klein E, Karry R, Sheinkman A, Kirsh Z, Mazor M, Tzukerman M, Ben-Shachar D. 2002. State-dependent alterations in mitochondrial complex I activity in platelets: a potential peripheral marker for schizophrenia. Mol Psychiatry 7(9):995-1001.
Abstract: Schizophrenia, the most severe psychiatric disorder, is characterized by heterogeneity of clinical signs, often categorized into positive and negative symptoms. Among a wide array of competing biological mechanisms, altered cerebral energy metabolism and mitochondrial dysfunction have been suggested to play an important role in the pathophysiology of schizophrenia. In this study we investigated mitochondrial complex I in platelets of 113 schizophrenic patients divided into three groups (acute psychotic episode, chronic active state and residual schizophrenia) and 37 control subjects. Complex I was analysed at the level of enzymatic activity, mRNA and protein levels by enzyme kinetics, RT-PCR and Western blot analyses, respectively. Complex I activity in platelets of schizophrenic patients altered with disease state presenting high specificity and sensitivity. Thus, increased activity was associated with psychotic symptomology, while its decrease was observed in patients with residual schizophrenia. The relationship between the clinical state and complex I activity in schizophrenia was further supported by its

positive correlation with the severity of patients' positive symptoms assessed by clinical ratings. In addition, similar alterations were observed at the levels of mRNA and protein of the 24- and 51-kDa iron-sulfur flavoprotein subunits of the complex. Taken together these results point to the potential of platelet complex I to turn into a reliable novel marker for schizophrenia. At present, definitive diagnosis depends only on descriptive behavioral and symptomatic information, therefore a peripheral measurable specific marker will contribute to diagnosis and monitoring of the disease.

Double KL, Ben-Shachar D, Youdim MBH, Zecca L, Riederer P, Gerlach M. 2002. Influence of neuromelanin on oxidative pathways within the human substantia nigra. Neurotoxicol Teratol 24(5):621-628.
Abstract: Neuromelanin (NM) is a dark-coloured pigment produced in the dopaminergic neurons of the human substantia nigra (SN). The function of NM within the pigmented neurons is unknown but other melanins are believed to play a protective role via attenuation of free radical damage. Experimental evidence suggests that NM may also exhibit this characteristic, possibly by directly inactivating free radical species or via its ability to chelate transition metals, such as iron. Increased tissue iron, however, may saturate iron-chelating sites on NM and a looser association between iron and NM may result in an increased, rather than decreased, production of free radical species. The death of NM-pigmented neurons in Parkinson's disease (PD) is associated with both a measurable increase in tissue iron concentrations and indices of free radical mediated damage, suggesting that NM is involved in the aetiology of this disorder. As yet, it is unknown whether NM in the parkinsonian brain differs to that found in healthy tissue and thus may fulfil a different role within this tissue. (C) 2002 Elsevier Science Inc. All rights reserved.

Crompton DE, Chinnery PF, Fey C, Curtis ARJ, Morris CM, Kierstan J, Burt A, Young F, Coulthard A, Curtis A, Ince PG, Bates D, Jackson MJ, Burn J. 2002. Neuroferritinopathy: A window on the role of iron in neurodegeneration. Blood Cells Mol Dis 29(3):522-531.
Abstract: Neuroferritinopathy is a recently recognised genetic disease resulting in a dominantly inherited movement disorder. The condition was mapped by linkage analysis to chromosome 19q13.3 and found to be due to a single adenine insertion in the ferritin light chain (FTL) gene at position 460-461 which is predicted to alter the C terminus of the FTL polypeptide. Clinical features of neuroferritinopathy are highly variable, with chorea, dystonia, and Parkinsonian features predominating in different affected individuals. The most consistent feature is a dystonic dysarthria. Symptoms and abnormal physical signs appear to be restricted to the nervous system and onset is typically in the fourth to sixth decades. Low serum ferritin also characterises this condition. Brain MR imaging of affected patients demonstrates iron deposition in the basal ganglia, progressing over years to cystic degeneration, and brain histochemistry shows abnormal aggregates of ferritin and iron. Now that the molecular basis of the condition is known, therapeutic interventions to reduce or reverse brain iron deposition are being evaluated. This rare disease provides evidence of a central role for iron metabolism in neurodegenerative disorders. (C) 2002 Elsevier Science (USA).

Crichton RR, Florence A, Ward RJ. 2002. Aluminium and iron in the brain - prospects for chelation. Coordination Chemistry Reviews 228(2):365-371.
Abstract: Aluminium and iron both accumulate in the brain in the course of ageing. We first briefly review how aluminium may interfere with iron metabolism through its interaction with iron homeostatic mechanisms. Then we present comparative data on the chelation of brain aluminium and iron in appropriate animal models of loading with the two metals. With both desferrioxamine (DFO) B and hydroxypyridone derivatives, brain iron is much more difficult to chelate than brain aluminium. This probably reflects the localisation of the former in ferritin and haemosiderin, and the latter in a more labile, non-protein form. The potential of long-term chelation therapy in the prevention of a number of neurodegenerative diseases

associated with ageing is discussed. (C) 2002 Elsevier Science B.V. All rights reserved.

Cordoba J, Sanpedro F, Alonso J, Rovira A. 2002. H-1 magnetic resonance in the study of hepatic encephalopathy in humans. Metab Brain Dis 17(4): 415-429.
Abstract: H-1 magnetic resonance ((HMR)-H-1) studies of the brain in patients with liver diseases have shown several abnormalities that may be relevant for the pathogenesis of hepatic encephalopathy. H-1 magnetic resonance imaging shows a typical pallidal hyperintensity on T-1-weighted images. This abnormality appears to be secondary to the accumulation of manganese in basal ganglia because of portal-systemic shunting. No direct correlation between the magnitude of pallidal hyperintensity and the grade of hepatic encephalopathy has been found, but some studies have related pallidal hyperintensity to parkinsonism. H-1 magnetic resonance spectroscopy shows relative to creatine an increase in glutamine/ glutamate (Glx) signal and a decrease of choline containing compounds (Cho) and myo-inositol. Abnormalities in the Glx signal have been interpreted as an increase in brain glutamine secondary to the metabolism of ammonia in astrocytes. Disturbances of Cho and myo-inositol have been interpreted as a compensatory response to the increase in intracellular osmolality caused by the accumulation of glutamine in astrocytes. In addition, magnetization transfer imaging shows signs compatible with low-grade cerebral edema. Altogether, (HMR)-H-1 studies suggest the accumulation of manganese and the development of osmotic abnormalities in the brain of patients with cirrhosis. These abnormalities appear to participate in some of the neurological manifestations of hepatic encephalopathy.

Commissaris Dacm, Nieuwenhuijzen Phja, Overeem S, De Vos A, Duysens JEJ, Bloem BR. 2002. Dynamic posturography using a new movable multidirectional platform driven by gravity. J Neurosci Methods 113(1): 73-84.
Abstract: Human upright balance control can be quantified using movable platforms driven by servo-controlled torque motors (dynamic posturography). We introduce a new movable platform driven by the force of gravity acting upon the platform and the subject standing on it. The platform consists of a 1 m(2) metal plate, supported at each of its four corners by a cable and two magnets. Sudden release of the magnets on three sides of the platform (leaving one side attached) induces rotational perturbations in either the pitch or roll plane. Release of all magnets causes a purely vertical displacement. By varying the slack in the supporting cables, the platform can generate small (0.5degrees) to very destabilising (19degrees) rotations. Experiments in healthy subjects showed that the platform generated standardised and reproducible perturbations, The peak rotation velocity well exceeded the threshold required to elicit postural responses in the leg muscles. Onset latencies were comparable to those evoked by torque motor-driven platforms. Randomly mixed multidirectional perturbations of large amplitude forced the subject to use compensatory steps (easily possible on the large support surface), with little confounding influence of habituation. We conclude that this gravity-driven multidirectional platform provides a useful and versatile tool for dynamic posturography. (C) 2002 Elsevier Science B.V. All rights reserved.

Collins MA, Neafsey EJ. 2002. Potential neurotoxic "agents provocateurs" in Parkinson's disease. Neurotoxicol Teratol 24(5):571-577.
Abstract: Idiopathic Parkinson's disease (PD), one of the most common neurodegenerative disorders associated with aging, is characterized neurochemically by abnormal and profound loss of nigrostriatal dopamine (DA) neurons. A prominent current view is that the excessive degeneration of the dopaminergic system is the outcome of extended insults by environmental neurotoxins or endogenous neurotoxic factors in genetically vulnerable or susceptible individuals. Recent insights into the identities and mechanisms of potential neurotoxic species, which span pesticides,

environmental contaminants including heterocyclic amines with beta-carboline (betaC) and isoquinoline (IQ) structures, endogenous DA metabolites or intermediates, neuromelanin, metals, and infectious agents, are presented. (C) 2002 Elsevier Science Inc. All rights reserved.

Chenais B, Morjani H, Drapier JC. 2002. Impact of endogenous nitric oxide on microglial cell energy metabolism and labile iron pool. J Neurochem 81(3): 615-623.
Abstract: Microglial activation is common in several neurodegenerative disorders. In the present study, we used the murine BV-2 microglial cell line stimulated with gamma-interferon and lipopolysaccharide to gain new insights into the effects of endogenously produced NO on mitochondrial respiratory capacity, iron regulatory protein activity, and redox-active iron level. Using polarographic measurement of respiration of both intact and digitonin-permeabilized cells, and spectrophotometric determination of individual respiratory chain complex activity, we showed that in addition to the reversible inhibition of cytochrome-c oxidase, long-term endogenous NO production reduced complex-I and complex-II activities in an irreversible manner. As a consequence, the cellular ATP level was decreased in NO-producing cells, whereas ATPase activity was unaffected. We show that NO up-regulates RNA-binding of iron regulatory protein 1 in microglial cells, and strongly reduces the labile iron pool. Together these results point to a contribution of NO derived from inflammatory microglia to the misregulation of energy-producing reactions and iron metabolism, often associated with the pathogenesis of neurodegenerative disorders.

Chen CJ, Liao SL. 2002. Oxidative stress involves in astrocytic alterations induced by manganese. Exp Neurol 175(1):216-225.
Abstract: It is hypothesized that manganese neurotoxicity could be secondary to a diminuition of cellular protective and scavenger mechanisms. Since manganese is known to be sequestered in glial cells, we investigated possible neurotoxic mechanisms involving astrocytes in vitro. Astrocytes differentiated into process-bearing stellate cells in response to manganese treatment. Manganese concentration dependently decreased cellular DNA synthesis, glial fibrillary acidic protein expression, energy production, antioxidant capacity, and glutamate transporter activity. In contrast, manganese increased glutamine synthetase protein expression and cytokine-stimulated interleukin 6 mRNA expression. Under the concentration of 0.1 mM manganese chloride caused no significant astrocyte death even up to 48 h after treatment. That is, these astrocytic alterations proceeded before the onset of cell demise. As a possible mediator of manganese-derived alterations, we determined intracellular redox state in astrocytes. Manganese time-dependently changed intracellular redox potential into oxidized state. The influx of manganese and its resultant oxidative stress was essential to most of the alterations, except for the action on stellation. Astrocytes are central component of the brain's antioxidant defense. Therefore, the observations suggest that dysfunction of astrocytes possibly involved in neurotoxic action of Manganese. (C) 2002 Elsevier Science (USA).

Castellani RJ, Perry G, Siedlak SL, Nunomura A, Shimohama S, Zhang J, Montine T, Sayre LM, Smith MA. 2002. Hydroxynonenal adducts indicate a role for lipid peroxidation in neocortical and brainstem Lewy bodies in humans. Neurosci Lett 319(1):25-28.
Abstract: Multiple lines of evidence indicate that oxidative stress is a critical pathogenic factor in Parkinson disease (PD) and diffuse Lewy body disease (DLBD). Previously, we demonstrated increased levels of redox-active iron in Lewy bodies, and that Lewy bodies accumulate advanced glycation end-products. To further characterize the role of oxidative stress in diseases with Lewy body formation, we examined immunocytochemically eight cases of PD and five cases of DLBD for adducts of the lipid peroxidation adduct 4-hydroxy-2-nonenal, and for N-epsilon-(carboxymethyl) lysine (CIVIL). Our findings demonstrate immunolocalization of 4-hydroxynonenal and CML to Lewy bodies in PD and DLBD. These findings not only support prior studies indicating that lipid peroxidation is increased in patients with

PD and DLBD but that oxidative damage may play a critical role in Lewy body formation. (C) 2002 Elsevier Science Ireland Ltd. All rights reserved.

Castaneda-Reyna MA, Ubilluz R, Avalos C, Escalante D. 2002. Wilson's disease: A clinical case is presented with neuropsychiatric dominant form and probable physiopathology interpretation with magnetic resonance of the brain. Rev Neurol 34(8):745-750.
Abstract: Case report. A clinical case of Wilson's disease is presented. She was a 26 years-old woman who began to show psychological symptoms, and later developed neurological signs such as asymmetrical hand tremor, parkinsonism, dystonia and later dysphagia and mutism, The ophthalmological examination found a Kayser-Fleischer ring in the Descemet membrane. There was disturbance of copper metabolism documented with reduction of serum ceruloplasmin and increase of the urinary excretion of copper. Cirrhosis was demonstrated through laparoscopy and liver biopsy. Results. The brain magnetic resonance showed frontotemporal atrophy and a degenerative process at the basal ganglia, cerebellum and brain stern, data which could de used to suggest the probable neuropsychiatric physiopathology. The stenosis and intense cervical dysphagia, associated with crycopharyngeal membrane, has not been mentioned previously.

Calabrese V, Scapagnini G, Ravagna A, Fariello RG, Stella AMG, Abraham NG. 2002. Regional distribution of heme oxygenase, HSP70, and glutathione in brain: Relevance for endogenous oxidant/antioxidant balance and stress tolerance. J Neurosci Res 68(1):65-75.
Abstract: It is generally recognized that lipid peroxides play an important role in the pathogenesis of several diseases and that sulfhydryl groups are critically involved in cellular defense against endogenous or exogenous oxidants. Recent evidence indicates that lipid peroxides directly participate in induction of cytoprotective proteins, such as heat shock proteins (Hsps), which play a central role in the cellular mechanisms of stress tolerance. Heme oxygenase (HO) is a stress protein that has been implicated in defense mechanisms against agents that may induce oxidative injury, such as endotoxins, cytokines and heme and its induction represents a common feature in a number of neurodegenerative diseases. In the present report we studied regional distribution of heme oxygenase (HO) activity and protein expression, together with that of Hps70, in brain of C57BL6 mice. Endogenous lipid peroxidation was investigated on the basis of the analysis of ultra weak chemiluminescence, hydro peroxides and lipid soluble fluorescent products, and compared to the regional distribution of thiols, antioxidant enzymes and trace metals. Our results show that levels of HO activity and expression of inducible Hsp70 and the ratio of GSH/GSSG in the different brain regions examined were positively correlated with the content of peroxides. Substantia Nigra was the brain area exhibiting the highest levels of HO-2, constitutive and inducible Hsp70, GSSG, peroxides, iron, and calcium, in contrast with the lowest content in GSH, GSH/GSSG ratio and glutathione reductase activity, compared to the other cerebral regions examined. Among these, cortex showed the lowest levels of HO-2, Hsp70, GSSG and peroxides that were associated with the highest levels of GSH and GSH/GSSG ratio. These data support the hypothesis that the glutathione redox state and basal peroxides can directly participate in the signaling pathways of heat shock protein expression and hence of stress tolerance. (C) 2002 Wiley-Liss, Inc.

Buzio L, De Palma G, Mozzoni P, Negrotti A, Scaglioni A, Calzetti S. 2002. Early onset of Parkinson's disease among subjects professionally exposed to either solvents or metals. Mov Disord 17:S144.

Burn DJ. 2002. Beyond the iron mask: Towards better recognition and treatment of depression associated with Parkinson's disease. Mov Disord 17(3): 445-454.
Abstract: This review examines the frequency of depression complicating Parkinson's disease (PD), its aetiology and clinical features, and also how it may be recognised and treated. Studies investigating the frequency of

depression in PD have yielded figures ranging between 2.7% and 70%. Methodological differences account for much of the disparity. The aetiology of depression in PD is complex, and probably relates to both biological and exogenous factors. Dysfunction of multiple neurotransmitter systems, including the serotonergic system, may be involved. Mood disturbances resulting from deep brain stimulation of the subthalamic nucleus may provide a fruitful area for future research, and assist our understanding of the neural networks involved in mediating depression. Several recent studies have confirmed that depression in the PD patient is a major determinant of quality of life and that this is closely related to dysfunction in other clinically important health areas. The validity for many existing scales in the screening, diagnosis, and monitoring of depression in the PD patient has not been established. The Montgomery-Asberg Depression Rating Scale and the Hamilton Rating Scale for Depression appear to have good diagnostic sensitivity and specificity when compared with DSM-IV criteria. Recommendations for the optimal drug treatment of depression in PD are difficult to give, due to an inexplicable dearth of sizeable, placebo-controlled studies. A majority of physicians would probably now opt for a selective serotonin reuptake inhibitor in the depressed PD patient. There is no good evidence that these drugs are associated with a worsening of motor features, but they should probably not be coprescribed with selegiline, because of the risk of causing a potentially serious serotonin syndrome. Several studies have suggested that depression in the PD patient is associated with a more rapid deterioration in cognitive and motor functions, perhaps as a surrogate marker for more extensive brainstem cell loss. (C) 2002 Movement Disorder Society.

Buchanan DD, Silburn PA, Chalk JB, Le Couteur DG, Mellick GD. 2002. The Cys282Tyr polymorphism in the HFE gene in Australian Parkinson's disease patients. Neurosci Lett 327(2):91-94.
Abstract: Iron homeostasis is altered in Parkinson's disease (PD). The HFE protein is an important regulator of cellular iron homeostasis and variations within this gene can result in iron overload and the disorder known as hereditary haemochromatosis. We studied the Cys282Tyr single nucleotide polymorphism as a genetic risk factor for PD in two distinct and separately collected cohorts of Australian PD patients and controls. In the combined cohort comprising 438 PD patients and 485 control subjects, we revealed an odds ratio for possession of the 282Tyr allele of 0.61 (95% confidence interval, Cl = 0.42-0.90, P = 0.011) from univariate chi-squared and 0.59 (95% Cl = 0.39-0.90, P = 0.014) after logistic regression analyses (correcting for potential confounding factors). These results suggest that possession of the 282Tyr allele may offer some protection against the development of PD. (C) 2002 Elsevier Science Ireland Ltd. All rights reserved.

Brodsky MA, Schilsky ML, Bronster DJ, Fatterpekar G, Naidich TP, Olanow W. 2002. Parkinsonism, manganese and brain imaging in liver failure. Mov Disord 17:S258.

Bounias M, Purdey M. 2002. Transmissible spongiform encephalopathies: a family of etiologically complex diseases - a review. Sci Total Environ 297(1-3): 1-19.
Abstract: The upsurge of 'mad cow disease' with its human implications has raised the problem of the etiological mechanisms and the similarities or differences underlying the family of transmissible spongiform encephalopathies. Structural properties of prions are reviewed in connection with their natural distribution and functions, factors of transmissibility and mechanisms of pathogenicity. Polymorphism is examined in relation to disease phenotype variants. The role of oxidative factors is emphasized, while raising complexity about the role of copper ions. Further investigation directions are suggested. (C) 2002 Elsevier Science B.V. All rights reserved.

Borie C, Gasparini F, Verpillat P, Bonnet AM, Agid Y, Hetet G, Brice A, Durr A, Grandchamp B. 2002. Association study between iron-related genes

polymorphisms and Parkinson's disease. J Neurol 249(7):801-804.
Abstract: We have conducted a case-control study in order to test for an association between 8 intragenic polymorphisms of 5 iron-related genes (transferrin, transferrin receptor I, HFE, frataxin and lactoferrin) and Parkinson disease. Comparison of genotypes and allele frequencies did not differ significantly between cases and controls for all studied polymorphisms except the G258S transferrin polymorphism, for which a higher frequency of the G allele was found among cases (p=0.033), particularly among, cases with onset older than 60 (p=0.0017) and with negative family history (p=0.022). This finding suggests that genetic variations in the control of iron metabolism may contribute to the pathogenesis of the disease.

Borae MF, Hadi SA. 2002. Respiratory chain enzyme activity and iron metabolism dysfunction in Parkinsonism. Eur Neuropsychopharmacol 12:S382.

Bolzoni F, Giraudo S, Lopiano L, Bergamasco B, Fasano M, Crippa PR. 2002. Magnetic investigations of human mesencephalic neuromelanin. Biochimica Et Biophysica Acta-Molecular Basis of Disease 1586(2):210-218.
Abstract: Pigmentation of neurons in substantia nigra is due to neuromelanin, a pigment that stores large amounts of iron. Human mesencephalic neuromelanin has been investigated by means of magnetic susceptibility measurements as 1 function of temperature. Magnetic measurements provide a physico-chemical characterization of the iron cluster buried in the organic melanin matrix and support the view that iron is not simply chelated, but rather is organized in a three-dimensional network. The paramagnetism of isolated iron ions is observed, in agreement with electron paramagnetic resonance spectroscopy. Furthermore, antiferromagnetic grains with a large size distribution function are present. These grains contain N spins coupled antiferromagnetically; however, N-1/2 spins are decoupled from the grain bulk and parallelly aligned. The latter subgrains are superparamagnetic with a blocking temperature ranging between 5 K and room temperature. This behavior has not been observed in synthetic melanin, where the paramagnetic contribution is strongly enhanced. Preliminary results on pigment isolated from patients affected by Parkinson's disease, a neurodegenerative pathology involving primarily pigmented neurons in substantia nigra pars compacta, show a lower total magnetization compared to control neuromelanin. The temperature behavior of zero field cooling and field cooling magnetizations is similar for both. The significant depletion of iron content in Parkinson's disease neuromelanin could indicate a progressive Fe migration from its storage environinent to the cytosol. (C) 2002 Elsevier Science B.V. All rights reserved.

Boland A, Gerardy J, Mossay D, Delapierre D, Seutin V. 2002. Pirlindole and dehydropirlindole protect rat cultured neuronal cells against oxidative stress-induced cell death through a mechanism unrelated to MAO-A inhibition. Br J Pharmacol 135(3):713-720.
Abstract: 1 It has been shown that the MAO (monoamine oxidase)-B inhibitor deprenyl (DPR, selegiline) protects some cell types against oxidative stress. By decreasing H2O2 production, MAO-A inhibitors could also reduce oxidative stress. 2 This study reports the effect of the MAO-A inhibitors, pirlindole (PIR), dehydropirlindole (DHP), brofaromine (BRO) and moclobemide (MCL) on primary-cultured brain cells exposed to iron-mediated toxicity. A comparison with trolox (TRO), a hydrosoluble vitamin-E analogue that protects against such an induced stress, was performed. 3 Rat hippocampal or cortical cultured cells were exposed either to 2 mum FeSO4 alone or in the presence of PIR, DHP, BRO, DPR, MCL or TRO. Cell survival (lactate-dehydrogenase measurements, 16 h incubation), intracellular peroxide production (DCF-fluorescence. I h incubation), lipoperoxidation (TBARS-fluorescence, 6 h incubation) and mitochondrial function (MTT-test, 16 h incubation) were assessed. 4 PIR, DHP and TRO significantly protected cultures ($P<0.05$) against Fe2+-induced toxicity in a concentration-dependent manner. The EC50s of these compounds were 6, 12 and 19 muM, respectively, in hippocampal cells. For cortical cell cultures

incubated in the presence of iron and PIR or DHP, EC50s were 5 and 6 muM respectively. All Hill coefficients were close to unity. BRO, MCL and DPR were not protective in any type of culture. The IC50s for the inhibition of MAO-A were 2, 2 and 0.2 muM for PIR, DHP and BRO, respectively. PIR, DHP and TRO, but not DPR, induced a significant decrease in both intracellular peroxide production and lipoperoxidation. They also improved mitochondrial function. 5 These experiments show that PIR and DHP can protect hippocampal and cortical neurons against oxidative stress at pharmacologically relevant concentrations. This protective effect seems unrelated to inhibition of MAO-A, but possibly involves free radical scavenging.

Biasetti M, Dawson R. 2002. Effects of sulfur containing amino acids on iron and nitric oxide stimulated catecholamine oxidation . Amino Acids 22(4): 351-368.
Abstract: Taurine is a free amino acid found in high concentrations in tissues containing catecholamines. The ability of taurine and its metabolic precursors to inhibit or stimulate catecholamine oxidation and subsequent quinone formation was examined. Ferric chloride was used as the catalyzing agent to stimulate L-dopa or norepinephrine oxidation and NO donors were also examined for their actions to stimulate quinone formation. Taurine attenuated iron-stimulated quinone formation from catecholamines suggesting that it may function as an endogenous antioxidant. Several other sulfur-containing amino acids (homocysteic acid, cysteine sulfinic acid and SAM) were found to inhibit catecholamine oxidation. Among other amino acids tested, homocysteine had biphasic effects; attenuating L-dopa oxidation catalyzed by ferric chloride and potentiating norepinephrine's oxidation catalyzed by both ferric chloride and sodium nitroprusside (SNP). Homotaurine and homocysteine (1 or 10mM) greatly stimulated SNP-induced norepinephrine oxidation. Homotaurine potentiated quinone formation in the presence of ferric iron and this effect was attenuated by desferroxamine. In order to exclude a possible NO/iron interaction in SNP's oxidizing action, SIN-1 chloride, a specific NO-donor, was tested as an oxidizing agent. The failure of desferroxamine or taurine to attenuate SIN-1 oxidation of norepinephrine suggests that peroxynitrite-mediated oxidation was likely the dominant mechanism. Our results show that endogenous sulfur containing amino acids, like taurine, could serve a protective role to reduce cellular damage associated with both NO and metal-stimulated catecholamine oxidation.

Bharath S, Hsu M, Kaur D, Rajagopalan S, Andersen JK. 2002. Glutathione, iron and Parkinson's disease. Biochem Pharmacol 64(5-6):1037-1048.
Abstract: Parkinson's disease (PD) is a progressive neurodegencrative disease involving neurodegeneration of dopaminergic neurons of the substantia nigra (SN), a part of the midbrain. Oxidative stress has been implicated to play a major role in the neuronal cell death associated with PD. Importantly, there is a drastic depletion in cytoplasmic levels of the thiol tripeptide glutathione within the SN of PD patients. Glutathione (GSH) exhibits several functions in the brain chiefly acting as an antioxidant and a redox regulator. GSH depletion has been shown to affect mitochondrial function probably via selective inhibition of mitochondrial complex I activity. An important biochemical feature of neurodegeneration during PD is the presence of abnormal protein aggregates present as intracytoplasmic inclusions called Lewy bodies. Oxidative damage via GSH depletion might also accelerate the build-up of defective proteins leading to cell death of SN dopaminergic neurons by impairing the ubiquitin-proteasome pathway of protein degradation. Replenishment of normal glutathione levels within the brain may hold an important key to therapeutics for PD. Several reports have suggested that iron accumulation in the SN patients might also contribute to oxidative stress during PD. (C) 2002 Elsevier Science Inc. All rights reserved.

Berg D, Roggendorf W, Schroder U, Klein R, Tatschner T, Benz P, Tucha O, Preier M, Lange KW, Reiners K, Gerlach M, Becker G. 2002. Echogenicity of the substantia nigra - Association with increased iron content and marker for

susceptibility to nigrostriatal injury. Arch Neurol 59(6):999-1005 .
Abstract: Background: Patients with Parkinson disease characteristically exhibit an increased echogenicity of the substantia nigra (SN) on transcranial sonography, a new neuroimaging technique. The same echo feature of the SN can be identified in 9% of healthy adults. Objective: To evaluate the relevance of the echogenic SN in healthy adults. Design: In the first part of the study, 10 healthy subjects younger than 40 years with a distinct SN hyperechogenicity underwent extensive neurological, motor, neuropsychological, and fluorine 18-dopa positron emission tomographic ([F-18]-dopa PET) examinations. Results were compared with those of 10 subjects with a low echogenic SN. In the second part of the study, the postmortem brains of 20 patients without extrapyramidal disorders during their lifetime were sonographically examined with a particular focus on SN echogenicity. Subsequently, one half of the brain was prepared for heavy metal analysis, the other for a histological examination. Results: Healthy subjects with SN hyperechogenicity exhibited a significant reduction of the [F-18] -dopa uptake, especially in the putamen (Wilcoxon matched pair test: left side, P=.006; right side, P=.009), whereas their neuropsychological and motor performance were normal. Postmortem studies showed that the echogenicity of the SN correlated with its iron content. Conclusions: Increased echogenicity of the SN, characteristically seen in Parkinson disease, is related to a functional impairment of the nigrostriatal system (even in young healthy adults) that can be revealed by [F-18]dopa PET studies. Substantia nigra hyperechogenicity is related to a higher tissue iron level, which is known to enhance the cells' generation of reactive oxygen specimens. Therefore, we hypothesize that transcranial sonography may identify a susceptibility marker for the development of nigral injury that can be detected early in life, prior to the onset of Parkinson disease.

Berg D, Gerlach M, Youdim MBH, Double KL, Zecca L, Riederer P, Becker G. 2002. Brain iron pathways and their relevance to Parkinson's disease (vol 79, pg 225, 2001). J Neurochem 80(4):719.

Arlt S, Beisiegel U, Kontush A. 2002. Lipid peroxidation in neurodegeneration: new insights into Alzheimer's disease. Curr Opin Lipidol 13(3):289-294.
Abstract: Imbalances of oxidative homeostasis and lipid peroxidation have been revealed as important factors involved in neurodegenerative disorders such as Alzheimer's disease. The brains of patients with Alzheimer's disease contain increased levels of lipid-peroxidation products such as 4-hydroxy-2-nonenal or acrolein, and enhanced lipid peroxidation can also be detected in cerebrospinal fluid and plasma from such patients. Recent research revealed that the interplay of transition metals, amyloid-beta peptide and lipid peroxidation might be responsible for increased oxidative stress and cell damage in this disease. In particular, the contrasting roles of amyloid-beta peptide, as a possible transition metal-chelating antioxidant for lipoproteins and a pro-oxidant when aggregated in brain tissue, has been the focus of discussion recently. In this context, lipid peroxidation has to be seen as an important part of the pathophysiological cascade in Alzheimer's disease, and its measurement in body fluids might serve as a therapy control for Alzheimer's disease and other neurodegenerative diseases. Curr Opin Lipidol 13:289-294. (C) 2002 Lippincott Williams Wilkins.

Aouad F, Florence A, Zhang Y, Collins F, Henry C, Ward RJ, Crichton RR. 2002. Evaluation of new iron chelators and their therapeutic potential. Inorganica Chimica Acta 339:470-480.
Abstract: One of the most important challenges for biological inorganic chemistry is to understand metal ion homeostasis, particularly since we are becoming increasingly aware that its disequilibrium is at the basis of a large number of clinical disorders. It is, therefore, also an important objective of our research to seek solutions, which can alleviate the consequences of the perturbation of the homeostasis of key metal ions within cells, tissues and organisms. Iron is one of the key metals in biology, for which disorders of homeostasis are implicated in a vast

panoply of pathological conditions. We review here the ways in which potential iron chelators can be evaluated, using appropriate in vivo or in vitro screening systems, and discuss their potential applications in the chelation of both iron and aluminium in the treatment of a number of human diseases. (C) 2002 Elsevier Science B.V. All rights reserved.

Anitha S, Shanmugavelu P, Gazula VR, Shankar SK, Menon RB, Rao RV, Rao JKS, Zecca L. 2002. Molecular Understanding of Aluminum Bioinorganic Chemistry in Relevance to the Pathology of Alzheimer's DiseaseVolume 822. p 228-245. Group 13 Chemistry: From Fundamentals to Applications: Acs Symposium Series.
Abstract: Aluminum (Al) is a suspected etiological factor in neurological disorders like Alzheimer's, Parkinson's, Huntington's diseases etc. The understanding of Al neurobiochemistry was hampered due to Al speciation chemistry and differential sensitivity animal models for Al toxicity. Experimental and circumstantial evidence provided a great deal of information on the complex inorganic biochemistry of Al in relevance to pathological events observed in Alzheimer's brains. In this contribution, the speciation chemistry of Al in relevance to neurobiology, role of Al, in modulating trace elemental homeostasis in human brains, Al-induced changes in animal brains mimicking Alzheimer's human brains, and its interaction with DNA are discussed.

Angel I, Bar A, Horovitz T, Taler G, Krakovsky M, Resnitsky D, Rosenberg G, Striem S, Friedman JE, Kozak A. 2002. Metal ion chelation in neurodegenerative disorders. Drug Development Research 56(3):300-309.
Abstract: Disturbance in metallochemical reactions and metal-protein association are associated with chronic neurodegenerative conditions, such as Alzheimer's and Parkinson's disease, as well as with neurodegeneration triggered by acute cerebral ischaemia. Many neurological diseases have been linked directly or indirectly to perturbed homeostasis of Ca, Fe, Zn, or Cu ions. Consequently, acute or chronic neurodegenerative disorders represent excellent targets for exploiting metal ion chelator approaches.

Anantharam V, Kitazawa M, Wagner J, Kaul S, Kanthasamy AG. 2002. Caspase-3-dependent proteolytic cleavage of protein kinase C delta is essential for oxidative stress-mediated dopaminergic cell death after exposure to methylcyclopentadienyl manganese tricarbonyl. J Neurosci 22 (5):1738-1751.
Abstract: In the present study, we characterized oxidative stress-dependent cellular events in dopaminergic cells after exposure to an organic form of manganese compound, methylcyclopentadienyl manganese tricarbonyl (MMT). In pheochromocytoma cells, MMT exposure resulted in rapid increase in generation of reactive oxygen species (ROS) within 5-15 min, followed by release of mitochondrial cytochrome C into cytoplasm and subsequent activation of cysteine proteases, caspase-9 (twofold to threefold) and caspase-3 (15- to 25-fold), but not caspase-8, in a time- and dose-dependent manner. Interestingly, we also found that MMT exposure induces a time- and dose-dependent proteolytic cleavage of native protein kinase Cdelta (PKCdelta, 72-74 kDa) to yield 41 kDa catalytically active and 38 kDa regulatory fragments. Pretreatment with caspase inhibitors (Z-DEVD-FMK or Z-VAD-FMK) blocked MMT-induced proteolytic cleavage of PKCdelta, indicating that cleavage is mediated by caspase-3. Furthermore, inhibition of PKCdelta activity with a specific inhibitor, rottlerin, significantly inhibited caspase-3 activation in a dose-dependent manner along with a reduction in PKCdelta cleavage products, indicating a possible positive feedback activation of caspase-3 activity by PKCdelta. The presence of such a positive feedback loop was also confirmed by delivering the catalytically active PKCdelta fragment. Attenuation of ROS generation, caspase-3 activation, and PKCdelta activity before MMT treatment almost completely suppressed DNA fragmentation. Additionally, overexpression of catalytically inactive PKCdelta(K376R) (dominant-negative mutant) prevented MMT-induced apoptosis in immortalized mesencephalic dopaminergic cells. For the first time, these data demonstrate that caspase-3-dependent proteolytic activation of

PKCdelta plays a key role in oxidative stress-mediated apoptosis in dopaminergic cells after exposure to an environmental neurotoxic agent.

Ramsden DB, Parsons RB, Ho SL, Waring RH. 2001 Dec. The aetiology of idiopathic Parkinson's disease. Mol Pathol 54(6):369-80.
Abstract: Agents potentially involved in the aetiology of idiopathic Parkinson's disease are discussed. These include factors regulating dopaminergic neurogenesis (Nurr 1, Ptx-3, and Lmx1b) and related proteins, together with genes involved in familial Parkinson's disease (alpha synuclein, parkin, and ubiquitin carboxy terminal hydroxylase L1), and endogenous and environmental agents.

Jiang H, Chen WF, Xie JX. 2001 Oct. [Relationship between dopamine and iron contents in the brain of parkinsonian rats]. Sheng Li Xue Bao 53(5):334-8.
Abstract: Using fast cyclic voltammetry (FCV), atomic absorption/flame emission spectrophotometry and high performance liquid chromatography for electrochemical detection, we studied the change in iron content in the substantia nigra (SN) of 6-hydroxydopamine (6-OHDA) lesioned Parkinsonian (PD) rats and the toxic effect of intranigral injection of iron on DA neurons. The neuroprotective effect of desferrioxamine mesylate was also observed. The results are as follows. (1) The iron content in SN on the lesioned side of 6-OHDA-lesioned PD rats was about three times as high as that in nonstandard PD rats. (2) The iron content in caudate putamen (CPu) on the lesioned side of PD rats was not different from that on the unlesioned side. (3) DA release as well as the content of DA and its metabolites were significantly decreased on the lesioned side of PD rats. (4) In the rats pretreated with intracerebroventricular desferrioxamine mesylate before 6-OHDA injection, the release and content of DA on the lesioned side were not significantly different from those on the unlesioned side. (5) Intranigral injection of 40 micrograms FeCl3 resulted in a dramatic reduction of both DA release and content in CPu. The above results strongly suggest that 6-OHDA reduces the DA release from CPu, in which iron plays an important role. Elevation of iron content in SN is one of the mechanisms responsible for the reduction of DA content. Desferrioxamine mesylate may exert a protective action on dopaminergic neurons.

Youdim MB. 2001 Aug. Deficiency and excess of iron in brain function and dysfunction. Nutr Rev 59(8 Pt 2):S83-5; discussion S85-7.

Paris I, Dagnino-Subiabre A, Marcelain K, Bennett LB, Caviedes P, Caviedes R, Azar CO, Segura-Aguilar J. 2001 Apr. Copper neurotoxicity is dependent on dopamine-mediated copper uptake and one-electron reduction of aminochrome in a rat substantia nigra neuronal cell line. J Neurochem 77 (2):519-29.
Abstract: The mechanism of copper (Cu) neurotoxicity was studied in the RCSN-3 neuronal dopaminergic cell line, derived from substantia nigra of an adult rat. The formation of a Cu-dopamine complex was accompanied by oxidation of dopamine to aminochrome. We found that the Cu-dopamine complex mediates the uptake of (64)CuSO(4) into the Raul Caviedes substantia nigra-clone 3 (RCSN3) cells, and it is inhibited by the addition of excess dopamine (2 m M) (63%, $p < 0.001$) and nomifensine (2 microM) (77%, $p < 0.001$). Copper sulfate (1 m M) alone was not toxic to RCSN-3 cells, but was when combined with dopamine or with dicoumarol (95% toxicity; $p < 0.001$) which inhibits DPNH and TPNH (DT)-diaphorase. Electron spin resonance (ESR) spectrum of the 5,5-dimethylpyrroline-N-oxide (DMPO) spin trap adducts showed the presence of a C-centered radical when incubating cells with dopamine, CuSO(4) and dicoumarol. A decrease in the expression of CuZn-superoxide dismutase and glutathione peroxidase mRNA was observed when RCSN-3 cells were treated with CuSO(4), dopamine, or CuSO(4) and dopamine. However, the mRNA expression of glutathione peroxidase remained at control levels when the cells were treated with CuSO(4), dopamine and dicoumarol. The regulation of catalase was different since all the treatments with CuSO(4) increased the expression of catalase mRNA. Our results suggest that copper

neurotoxicity is dependent on: (i) the formation of Cu-dopamine complexes with concomitant dopamine oxidation to aminochrome; (ii) dopamine-dependent Cu uptake; and (iii) one-electron reduction of aminochrome.

Obata T, Yamanaka Y. 2001 Feb. [Parkinsonism induced by MPTP and free radical generation]. Nippon Yakurigaku Zasshi 117(2):105-10.
Abstract: Oxygen free radical formation has been implicated in dopaminergic toxicity caused by 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) and iron. Although MPTP produces a parkinsonian syndrome after its conversion to 1-methyl-4-phenylpyridine (MPP+) by type B monoamine oxidase (MAO-B) in the brain, the etiology of this disease remains obscure. MPP+ is one of the most potent dopamine (DA)-releasing agents. Iron-catalyzed DA autoxidation and oxidative stress may be involved in the pathogenesis of Parkinson's disease. If indeed the effect of MPP+ on hydroxyl radical (.OH) formation is due to DA release, reserpine-induced DA depletion may reduce MPP(+)-induced .OH formation. Imidapril, an angiotensin converting enzyme (ACE) inhibitor, can resist MPP(+)-induced .OH formation via suppression of release of DA by angiotensin. Histidine, a singlet oxygen (1O2) scavenger, protects MPP (+)-induced .OH formation. Fluvastatin, an inhibitor of low-density lipoprotein (LDL) oxidation, can resist MPP(+)-induced .OH formation. The inhibitory effect on the susceptibility of LDL oxidation can reduce .OH generation. These drugs may be applied as antiparkinsonian agents. Further clinical investigation is necessary in the future.

Larumbe Ilundain R, Ferrer Valls JV, Vines Rueda JJ, Guerrero D, Fraile P. 2001 Jan-Feb. [Case-control study of markers of oxidative stress and metabolism of blood iron in Parkinson's disease]. Rev Esp Salud Publica 75 (1):43-53.
Abstract: BACKGROUND: Increasingly growing evidence exists of the involvement of oxidative stress mechanisms in Parkinson's disease. Lower levels of GSH in the sustantia nigra, an increase in iron buildup, an increase in the byproducts of lipid peroxidation and alterations in the mitochondrial complex I have been described. However, few studies have been made of levels of antioxidants in the peripheral bloodstream and of the influence of the intake of nutrients on the development of this disease. METHODS: In a group of 79 patients afflicted with idiopathic Parkinson's disease and a control group comprised of 107 subjects, compared by age, sex and place of residence, the lowered levels in the plasma of glutathione (GSH), malon dialdehyde (MDA), uric acid, tocophenol, beta-carotene, lycopene and different iron metabolism parameters were studied. Likewise, the intake of certain antioxidants was estimated based on a dietary survey. RESULTS: Significant differences ($p < or = 0.001$) were found in the plasma levels of GSH between cases (0.10 mumol/ml +/- 0.06) and controls (0.29 mumol/ml +/- 0.12). Likewise, the plasma levels of uric acid were lower ($p < or = 0.05$) in the cases (4.96 mg/ml +/- 1.96) than in the control groups (5.39 mg/ml +/- 1.13). No significant difference was found in the plasma levels of MDA, tocopherol, beta-carotene and lycopene. With regard to iron metabolism, significantly higher ferritine and transferrin values were found in the patients with EP than in the control group, showing a lower transferrin saturation percentage ($p < or = 0.05$). The iron showed no significant changes between cases and control groups. CONCLUSIONS: The results of this study support the possible involvement of oxidative stress in the pathogenesis of Parkinson's disease and reveal, in turn, alterations in some peripheral blood parameters in keeping with known findings in the sustantia nigra.

Zhou B, Westaway SK, Levinson B, Johnson MA, Gitschier J, Hayflick SJ. 2001. A novel pantothenate kinase gene (PANK2) is defective in Hallervorden-Spatz syndrome. Nat Genet 28(4):345-349.
Abstract: Hallervorden-Spatz syndrome (HSS) is an autosomal recessive neurodegenerative disorder associated with iron accumulation in the brain. Clinical features include extrapyramidal dysfunction, onset in childhood, and a relentlessly progressive course(1). Histologic study reveals iron deposits in the basal ganglia(2). In this respect, HSS may serve as a

model for complex neurodegenerative diseases, such as Parkinson disease (3), Alzheimer disease(4), Huntington diseases and human immunodeficiency virus (HIV) encephalopathy(6), in which pathologic accumulation of iron in the brain is also observed. Thus, understanding the biochemical defect in HSS may provide key insights into the regulation of iron metabolism and its perturbation in this and other neurodegenerative diseases. Here we show that HSS is caused by a defect in a novel pantothenate kinase gene and propose a mechanism for oxidative stress in the pathophysiology of the disease.

Zheng W, Zhao QQ. 2001. Iron overload following manganese exposure in cultured neuronal, but not neuroglial cells. Brain Res 897(1-2):175-179.
Abstract: Our previous studies show that manganese (Mn) exposure inhibits aconitase, an enzyme regulating the proteins responsible for cellular iron (Fe) equilibrium. This study was performed to investigate whether Mn intoxication leads to an altered cellular Fe homeostasis in cultured neuronal or neuroglial cells as a result of disrupted Fe regulation. Our results reveal a significant increase in the expression of transferrin receptor (TfR) mRNAs and a corresponding increase in cellular Fe-59 net uptake by PC12 cells, but not astrocytes, following Mn exposure. These findings suggest that alteration by Mn of cellular Fe homeostasis may contribute to Mn-induced neuronal cytotoxicity. (C) 2001 Elsevier Science B.V. All rights reserved.

Zheng W. 2001. Toxicology of choroid plexus: Special reference to metal-induced neurotoxicities. Microsc Res Tech 52(1):89-103.
Abstract: The chemical stability in the brain underlies normal human thinking, learning, and behavior. Compelling evidence demonstrates a definite capacity of the choroid plexus in sequestering toxic heavy metal and metalloid ions. As the integrity of blood-brain and blood-CSF barriers, both structurally and functionally, is essential to brain chemical stability, the role of the choroid plexus in metal-induced neurotoxicities has become an important, yet under-investigated research area in neurotoxicology. Metals acting on the choroid plexus can be categorized into three major groups. A general choroid plexus toxicant can directly damage the choroid plexus structure such as mercury and cadmium. A selective choroid plexus toxicant may impair specific plexus regulatory pathways that are critical to brain development and function, rather than induce massive pathological alteration. The typical examples in this category include lead-induced alteration in transthyretin production and secretion as well as manganese interaction with iron in the choroid plexus. Furthermore, a sequestered choroid plexus toxicant, such as iron, silver, or gold, may be sequestered by the choroid plexus as an essential CNS defense mechanism. Our current knowledge on the toxicological aspect of choroid plexus research is still incomplete. Thus, the future research needs have been suggested to focus on the role of choroid plexus in early CNS development as affected by metal sequestration in this tissue, to explore how metal accumulation alters the capacity of the choroid plexus in regulation of certain essential elements involved in the etiology of neurodegenerative diseases, and to better understand the blood-CSF barrier as a defense mechanism in overall CNS function. (C) 2001 Wiley-Liss, Inc.

Zheng W. 2001. Neurotoxicology of the brain barrier system: New implications. J Toxicol Clin Toxicol 39(7):711-719.
Abstract: The concept of a barrier system in the brain has existed for nearly a century. The barrier that separates the blood from the cerebral interstitial fluid is defined as the blood-brain barrier, while the one that discontinues the circulation between the blood and cerebrospinal fluid is named the blood-cerebrospinal fluid barrier. Evidence in the past decades suggests that brain barriers are subject to toxic insults from neurotoxic chemicals circulating in blood. The aging process and some disease states render barriers more vulnerable to insults arising inside and outside the barriers. The implication of brain barriers in certain neurodegenerative diseases is compelling, although the contribution of chemical-induced barrier dysfunction in the etiology, of any of these disorders remains

poorly understood. This review examines what is currently, understood about brain barrier systems in central nervous system disorders by focusing on chemical-induced neurotoxicities including those associated with nitrobenzenes, N-methyl-D-aspartate, cyclosporin A, pyridostigmine bromide, aluminum, lead, manganese, 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine, and 3-nitropropionic acid. Contemporary research questions arising from this growing understanding show enormous promises for brain researchers, toxicologists, and clinicians.

Zecca L, Tampellini D, Rizzio E, Giaveri G, Gallorini M. 2001. The determination of iron and other metals by INAA in Cortex, Cerebellum and Putamen of human brain and in their neuromelanins. Journal of Radioanalytical and Nuclear Chemistry 248(1):129-131.
Abstract: Neuromelanin (NM) is an ubiquitary intraneuronal pigment in human brain and its abundance is probably related to neuronal vulnerability, especially in Parkinson Disease. In this and other neurodegenerative diseases an association was shown with endogenous accumulation and environmental exposure to metals. Melanins are strong metal chelators, therefore it is important to measure their content in brain regions and in their NMs. In this work the concentration of 18 elements was measured in each brain region and in the corresponding NM by instrumental neutron activation analysis.

Zecca L, Tampellini D, Gerlach M, Riederer P, Fariello RG, Sulzer D. 2001. Substantia nigra neuromelanin: structure, synthesis, and molecular behaviour. Journal of Clinical Pathology-Molecular Pathology 54(6): 414-418.
Abstract: The pigmented neurones of the substantia nigra are typically lost in Parkinson's disease; however, the possible relation between neuronal vulnerability and the presence of neuromelanin has not been elucidated. Early histological studies revealed the presence of increasing amounts of neuromelanin in the substantia nigra with aging in higher mammals, showed that the neuromelanin granules are surrounded by a membrane, and comparatively evaluated the pigmentation of the substantia nigra in different animal species. Histochemical studies showed the association of neuromelanin with lipofuscins. However, systematic investigations of the structure, synthesis, and molecular interactions of neuromelanin have been undertaken only during the past decade. In these later studies, neuromelanin was identified as a genuine melanin with a strong chelating ability for iron and an affinity for compounds such as lipids, pesticides, and MPP+. The affinity of neuromelanin for a variety of inorganic and organic toxins is consistent with a postulated protective function for neuromelanin. Moreover, the neuronal accumulation of neuromelanin during aging and the link between its synthesis and a high cytosolic concentration of catechols suggest a protective role. However, its putative neuroprotective effects could be quenched in conditions of toxin overload.

Zecca L, Tampellini D, Costi P, Rizzio E, Giaveri G, Gallorini M. 2001. Combined biochemical separation and INAA for the determination of iron and other metals in Neuromelanin of human brain Substantia Nigra . Journal of Radioanalytical and Nuclear Chemistry 249(2):449-454.
Abstract: In Parkinson disease Fe and other metals increase in Substantia Nigra (SN) and other basal nuclei. Since Fe can generate citotoxic free radicals, Neuromelanin (NM) could play an important protective role in neurons. In this work an original procedure for separation of NM, preparation of samples and analysis is presented. The determination of SN and its NM elemental content was carried out by instrumental neutron activation analysis. Several actions were taken to reduce the metal contaminations: use of high purity reagents, dissection of tissues with titanium coated tools and adequate processing of samples.

Zecca L, Gallorini M, Schunemann V, Trautwein AX, Gerlach M, Riederer P, Vezzoni P, Tampellini D. 2001. Iron, neuromelanin and ferritin content in the substantia nigra of normal subjects at different ages: consequences for iron storage and neurodegenerative processes. J Neurochem 76(6):

1766-1773.
Abstract: Information on the molecular distribution and ageing trend of brain iron in post-mortem material from normal subjects is scarce. Because it is known that neuromelanin and ferritin form stable complexes with iron(III), in this study we measured the concentration of iron, ferritin and neuromelanin in substantia nigra from normal subjects, aged between 1 and 90 years, dissected post mortem. Iron levels in substantia nigra were 20 ng/mg in the first year of life, had increased to 200 ng/mg by the fourth decade and remained stable until 90 years of age. The H-ferritin concentration was also very low (29 ng/mg) during the first year of life but increased rapidly to values of approximate to 200 ng/mg at 20 years of age, which then remained constant until the eighth decade of life. L-Ferritin also showed an increasing trend during life although the concentrations were approximate to 50% less than that of H-ferritin at each age point. Neuromelanin was not detectable during the first year, increased to approximate to 1000 ng/mg in the second decade and then increased continuously to 3500 ng/mg in the 80th year. A Mossbauer study revealed that the high-spin trivalent iron is probably arranged in a ferritin-like iron-oxyhydroxide cluster form in the substantia nigra. Based on this data and on the low H- and L-ferritin content in neurones it is concluded that neuromelanin is the major iron storage in substantia nigra neurones in normal individuals.

Youdim MBH. 2001. Deficiency and excess of iron in brain function and dysfunction. Nutr Rev 59(8):S83-S85.

Yoshida S, Ektessabi A, Fujisawa S. 2001. XANES spectroscopy of a single neuron from a patient with Parkinson's disease. Journal of Synchrotron Radiation 8:998-1000.
Abstract: Chemical state of transition metals such as iron in a single neuron of the substantia nigra (SN) from a patient with Parkinson's disease (PD) was studied in this paper, using autopsy midbrain specimens including SN. Parkinson's disease is one of the major neurodegenerative diseases. The excessive accumulation of iron and its chemical states in SN neurons are related to the oxidative damage leading to neuronal cell death. X-ray absorption fine structure spectroscopy (XAFS) with SR micro beam (10 mum) was used to investigate the chemical state of iron in the SN neurons. X-ray absorption near-edge structure (XANES) spectroscopy results showed that the chemical state of iron in the neuromelanin granules within SN neurons changed from ferrous (Fe2+) to ferric (Fe3+) ion in the process of neuronal degeneration.

Yase Y, Yoshida S, Kihira T, Wakayama I, Komoto J. 2001. Kii ALS dementia. Neuropathology 21(2):105-109.
Abstract: Epidemiological surveys in the foci of ALS of the Kii Peninsula of Japan started in the early 1960s. Continuous surveys conducted for decades revealed that there have been two foci in the Kii Peninsula: one in Kozagawa in the southern part, and the other in Hobara in the south-east. Clinically, ALS patients of the Kii foci occasionally showed parkinsonian features or dementia that have not been reported in the sporadic form of ALS. Neuropathologically, numerous NFT that are identical to those of Alzheimer's disease were observed in the cerebral cortex and in the brainstem nuclei. To elucidate the etiopathogenesis of this unique form of ALS, an analysis was conducted of the environment in the focus areas and of the specimens from the patients with ALS. It was hypothesized that the long exposure of these environments to low calcium and magnesium, and an excess of aluminum and manganese in the drinking water and the soil, might lead to the deposition of some trace elements in the CNS, eventually causing neuronal degeneration and death.

Wissler JH, Logemann E. 2001. Proteins of neurodegenerative diseases scrapie [cellular prion, PrP], alzheimer [amyloid precursor, APP], parkinson [parkin, P] and huntington [huntingtin, H] are related in canonical [Cu/Zn]-metalloregulator and RNA-binding [R3H] domains. Biophys J 80(1):566A.

Winkler AS, Marsden J, Parton M, Watkins PJ, Chaudhuri KR. 2001. Erythropoietin deficiency and anaemia in multiple system atrophy. Mov Disord 16(2): 233-239.
Abstract: Serum erythropoietin (EPO) levels are partially controlled by the sympathetic outflow to the kidney. We have studied whether patients with multiple system atrophy (MSA), known to be associated with dysautonomia. are EPO-deficient. Eighteen MSA patients were studied along with 32 idiopathic Parkinson's disease (PD) patients, 23 controls with iron-deficiency anaemia, and 18 healthy individuals. Serum creatinine was normal in all groups. Mean haemoglobin (Hb) concentration in MSA patients was 13.7 +/- 1.7 g/dL. Four MSA patients had unexplained anaemia (minimum Hb: 10.5 g/dL) and abnormal autonomic function tests including significant postural hypotension, whereas none of the PD patients was anaemic. Serum EPO levels were suppressed in relation to anaemia in MSA patients compared to elevated EPO levels in iron-deficiency anaemia patients (difference of regression lines P < 0.001), indicating EPO deficiency in the anaemic MSA patients. Serum EPO levels in PD patients were within normal range. A subset of MSA patients has anaemia and postural hypotension. which may be associated with EPO deficiency. This may have therapeutic implications. <(c)> 2001 Movement Disorder Society.

Wang XF, Cynader MS. 2001. Pyruvate released by astrocytes protects neurons from copper-catalyzed cysteine neurotoxicity. J Neurosci 21(10): 3322-3331.
Abstract: We have found previously that astrocytes can provide cysteine to neurons. However, cysteine has been reported to be neurotoxic although it plays a pivotal role in regulating intracellular levels of glutathione, the major cellular antioxidant. Here, we show that cysteine toxicity is a result of hydroxyl radicals generated during cysteine autoxidation. Transition metal ions are candidates to catalyze this process. Copper substantially accelerates the autoxidation rate of cysteine even at submicromolar levels, whereas iron and other transition metal ions, including manganese, chromium, and zinc, are less efficient. The autoxidation rate of cysteine in rat CSF is equal to that observed in the presence of similar to0.2 muM copper. In tissue culture tests, we found that cysteine toxicity depends highly on its autoxidation rate and on the total amount of cysteine being oxidized, suggesting that the toxicity can be attributed to the free radicals produced from cysteine autoxidation, but not to cysteine itself. We have also explored the in vivo mechanisms that protect against cysteine toxicity. Catalase and pyruvate were each found to inhibit the production of hydroxyl radicals generated by cysteine autoxidation. In tissue culture, they both protected primary neurons against cysteine toxicity catalyzed by copper. This protection is attributed to their ability to react with hydrogen peroxide, preventing the formation of hydroxyl radicals. Pyruvate, but not catalase or glutathione peroxidase, was detected in astrocyte-conditioned medium and CSF. Our data therefore suggest that astrocytes can prevent cysteine toxicity by releasing pyruvate.

Villalobos V, Estevez J, Novo E, Bonilla E. 2001. Effects of chronic manganese treatment on mouse brain (H-3) spiroperidol binding parameters: In vivo and in vitro studies. Revista Cientifica-Facultad De Ciencias Veterinarias 11 (4):306-313.
Abstract: The in vivo and in vitro effects of Mn on the binding of (H-3) spiroperidol to mouse brain was assessed. (H-3) spiroperidol bind ing parameters (Kd and Bmax) in striatum, hypothalamus and olfactory bulb did not change by Mn administration (5mg/kg/day) for 9 weeks. On the other hand, preincubation of mouse brain homogenates with increasing concentrations of Mn (0.05-10 mM) and dopamine (10 mM) resulted in a significant rise in the (H-3) spiroperidol specific binding with a Mn concentration of 75 muM or higher. Binding assays carried out using homogenates preincubated with 10 mM dopamine and 75 muM Mn showed an increase in Bmax and Kd. These studies demonstrated that Mn administration does not after the binding pattern of (H-3) spiroperidol. The increase in Bmax and Kd observed in the in vitro assays, when dopamine

and Mn are added to the incubation medium seem to be originated from changes in cell membranes, leading to the exposure of new and different binding sites.

Vanacore N, Bonifati V, Fabbrini G, Colosimo C, De Michele G, Marconi R, Nicholl D, Locuratolo N, Talarico G, Romano S, Stocchi F, Bonuccelli U, De Mari M, Vieregge P, Meco G. 2001. Epidemiology of multiple system atrophy. ESGAP Consortium. European Study Group on Atypical Parkinsonisms. Neurological Sciences 22(1):97-99.
Abstract: Multiple system atrophy (MSA) is a form of atypical parkinsonism with unknown etiology. The epidemiological studies conducted up to now on this disease are scarce. The incidence rate is about 0.6 cases per 100 000 persons per year. The prevalence rates show 4-5 cases per 100 000 persons. In Italy, about 4900 prevalent cases have been estimated. The mean onset age is about 54 years; the median survival is 7-9 years. Only one case-control study has been performed on this disease. This study showed an increased risk of MSA associated with occupational exposure to organic solvents, plastic monomers and additives, pesticides and metals. Smoking habits seem to be less frequent in MSA cases (as in Parkinson's disease cases) than in healthy controls. Quinn's clinical criteria and those of the Consensus Conference promoted by the American Academy of Neurology are in fair agreement. We have performed a case-control study on 73 MSA cases, 146 hospital controls and 73 population controls.

Uversky VN, Li J, Fink AL. 2001. Metal-triggered structural transformations, aggregation, and fibrillation of human alpha-synuclein - A possible molecular link between Parkinson's disease and heavy metal exposure. J Biol Chem 276(47):44284-44296.
Abstract: Parkinson's disease involves the aggregation of alpha -synuclein to form fibrils, which are the major constituent of intracellular protein inclusions (Lewy bodies and Lewy neurites) in dopaminergic neurons of the substantia nigra. Occupational exposure to specific metals, especially manganese, copper, lead, iron, mercury, zinc, aluminum, appears to be a risk factor for Parkinson's disease based on epidemiological studies. Elevated levels of several of these metals have also been reported in the substantia nigra of Parkinson's disease subjects. We examined the effect of various metals on the kinetics of fibrillation of recombinant alpha -synuclein and in inducing conformational changes, as monitored by biophysical techniques. Several di- and trivalent metal ions caused significant accelerations in the rate of alpha -synuclein fibril format-ion. Aluminum was the most effective, along with copper(H), iron(III), cobalt (III), and manganese(II). The effectiveness correlated with increasing ion charge density. A correlation was noted between efficiency in stimulating fibrillation and inducing a conformational change, ascribed to formation of a partially folded intermediate. The potential for ligand bridging by polyvalent metal ions is proposed to be an important factor in the metal-induced conformational changes of a-synuclein. The results indicate that low concentrations of some metals can directly induce alpha -synuclein fibril formation.

Troadec JD, Marien M, Darios F, Hartmann A, Ruberg M, Colpaert F, Michel PP. 2001. Noradrenaline provides long-term protection to dopaminergic neurons by reducing oxidative stress. J Neurochem 79(1):200-210.
Abstract: To better understand the neurotrophic function of the neurotransmitter noradrenaline, we have developed a model of mesencephalic cultures in which we find low concentrations (0.3-10 muM) of noradrenaline to be remarkably effective in promoting long-term survival and function of dopaminergic neurons. This protective action reproduced the effect of caspase inhibition. It was atypical in that it occurred independently of adrenoceptor activation and was mimicked by some antioxidants, redox metal chelators and the hydroxyl radical detoxifying enzyme catalase. Interestingly, intracellular reactive oxygen species (ROS) were drastically reduced by treatment with noradrenaline, indicating that the neurotransmitter itself acted as an antioxidant. Prevention of oxidative stress was, however, independent of the

glutathione antioxidant defense system. Chemical analogues of noradrenaline bearing two free hydroxyl groups in the ortho position of the aromatic ring (o-catechols), as well as o-catechol itself, mimicked the survival promoting effects of the neurotransmitter, suggesting that this diphenolic structure was critical for both neuroprotection and reduction of ROS production. Paradoxically, the autoxidation of noradrenaline and the ensuing production of quinone metabolites may be required for both effects, as the neurotransmitter was spontaneously and rapidly degraded over time in the culture medium. These results support the concept that central noradrenergic mechanisms have a neuroprotective role, perhaps in part by reducing oxidative stress.

Tomkins J, Banner SJ, Mcdermott CJ, Shaw PJ. 2001. Mutation screening of manganese superoxide dismutase in amyotrophic lateral sclerosis. Neuroreport 12(11):2319-2322.
Abstract: Seventy-seven cases of ALS were screened for mutations in the manganese superoxide dismutase gene (SOD2). DNA was extracted from CNS tissue and screened using single stranded conformation polymorphism and heteroduplex analysis. No mutations were identified in the entire coding region of the SOD2 gene. The known polymorphism in the mitochondrial targeting sequence was identified. No association was found between this polymorphism and ALS. A further polymorphism was detected in the intronic sequence upstream of exon 4, though no association with ALS was demonstrated. We therefore conclude that mutations in SOD2 do not appear to cause ALS. NeuroReport 12:2319-2322 (C) 2001 Lippincott Williams & Wilkins.

Thompson KJ, Shoham S, Connor JR. 2001. Iron and neurodegenerative disorders. Brain Res Bull 55(2):155-164.
Abstract: The brain shares with other organs the need for a constant and readily available supply of iron and has a similar array of proteins available to it for iron transport, storage, and regulation. However, unlike other organs, the brain places demands on iron availability that are regional, cellular, and age sensitive. Failure to meet these demands for iron with an adequate supply in a timely manner can result in persistent neurological and cognitive dysfunction. Consequently, the brain has developed mechanisms to maintain a continuous supply of iron. However, in a number of common neurodegenerative disorders, there appears to be an excess accumulation of iron in the brain that suggests a loss of the homeostatic mechanisms responsible for regulating iron in the brain. These systems are reviewed in this article. As a result of a loss in iron homeostasis, the brain becomes vulnerable to iron-induced oxidative stress. Oxidative stress is a confounding variable in understanding the cell death that may result directly from a specific disease and is a contributing factor to the disease process. The underlying pathogenic event in oxidative stress is cellular iron mismanagement. (C) 2001 Elsevier Science Inc.

Takeda A, Takatsuka K, Connor JR, Oku N. 2001. Abnormal iron accumulation in the brain of neonatal hypotransferrinemic mice. Brain Res 912(2):154-161.
Abstract: Transferrin is a plasma protein involved in iron delivery to tissues. To study iron transport into the brain under a transferrin deficiency, iron concentration and Fe-59 uptake in the brain were measured in neonatal hypotransferrinemic (HP) mice at 7 days of age. Brain iron concentration of the HP mice, in which iron concentration was relatively high in the cerebral cortex and cerebellum, was approximately three times higher than that of non-mutant mice, whereas serum iron concentration of HP mice was significantly lower than that of non-mutant mice. When (FeCl)-Fe-59, was subcutaneously injected into HP and non-mutant mice, 59Fe was distributed highly in the choroid plexus in the ventricles of HP mice 24 h after injection. The Fe-59 distribution in the brain was different between HP and non-mutant mice. On the other hand, the clearance of Fe-59 from the blood was very high in HP mice and the hepatic Fe-59 concentration of HP mice was more than ten times of that of non-mutant mice. The present findings demonstrate that iron distribution in the brain is changed by transferrin deficiency and that iron abnormally

accumulates in the brain of HP mice. It is likely that the management of iron is different in the brain of HP mice. (C) 2001 Elsevier Science B.V. All rights reserved.

Takeda A. 2001. Significance of transferrin in iron delivery to the brain. Journal of Health Science 47(6):520-524.
Abstract: The role of transferrin in iron delivery to tissues is described. Transferrin-dependent iron uptake by erythroid cells in the bone marrow is essential Cor the development of erythrocytes, while nontransferrin-bound iron can be taken up in tissues such as liver. Oil the basis of the evidence that iron distribution in the body is changed by iron saturation of plasma transferrin. the role of transferrin in iron delivery to the brain is reviewed. In the case of transient iron saturation of plasma transferrin. Fe-59 concentrations in the brain of iron-loaded mice are approximately 40-50% of those of control mice in all brain regions tested except the choroid plexus. in which the Fe-59 concentration is equal. A similar distribution of Fe-59 in the brain is also observed in neonatal hypotransferrinemic (HP) mice, which have a splicing defect in the transferrin gene. resulting in < 1% of the normal plasma levels of transferrin. These results suggest that transferrin-bound iron is responsible Cor the fraction of iron in the circulation that enters the brain. On the other hand. the iron concentration in the brain of HP mice is approximately three times higher than that in nonmutant mice. It is likely that the management of iron is affected in the brain of HP mice. Brain transferrin may he involved in the management of iron in the brain.

Takanashi M, Mochizuki H, Yokomizo K, Hattori N, Mori H, Yamamura Y, Mizuno Y. 2001. Iron accumulation in the substantia nigra of autosomal recessive juvenile parkinsonism (ARJP). Parkinsonism & Related Disorders 7(4): 311-314.
Abstract: Autosomal recessive juvenile parkinsonism (ARJP) is a distinct clinical and genetic entity characterized by highly selective neuronal death in the substantia nigra (SN) and locus coeruleus neurons without Lewy body formation. The mechanism of neuronal death of ARJP is still unknown. Our study demonstrated that iron staining was more intense in ARJP than in both controls and sporadic Parkinson's disease (PD), and there were differences in the pattern of distribution of iron staining between ARJP and PD. In addition neurites of SN in ARJP showed intense iron staining. Thus, we postulate that oxidative stress may play an important role in the neurodegeneration that occurs in ARJP. (C) 2001 Elsevier Science Ltd. All rights reserved.

Takahashi S, Takahashi I, Sato H, Kubota Y, Yoshida S, Muramatsu Y. 2001. Age-related changes in the concentrations of major and trade elements in the brain of rats and mice. Biol Trace Elem Res 80(2):145-158.
Abstract: Age-related changes in the concentrations of constituent elements in the brains of rats and mice 1 wk to 24 mo old were determined with inductively coupled plasma-mass spectrometry (ICP-MS) and inductively coupled plasma-atomic emission spectrometry (ICP-AES). Seventeen elements could be determined with reasonable accuracy and reproducibility. They were P, K, Na, Mg, Ca, Fe, Zn, Cu, Rb, Al, Mn, Sr, Mo, Co, Pb, Cs, and Cd in order of concentrations in the adult rat brains. In these elements, six major elements (P, K, Na, Fe, Mg, Ca) were determined with ICP-AES and the others with ICP-MS. The concentrations of each element and the pattern of age-related changes were similar between the rat and mouse brains. The elements of which concentrations decreased with aging were K and Rb. On the other hand, the concentrations of some metal elements, including Fe, Cu, Sr, and Co, appeared to increase with growth and aging. The concentrations of other elements were relatively constant throughout the age examined.

Sunderman FW. 2001. Review: Nasal toxicity, carcinogenicity, and olfactory uptake of metals. Ann Clin Lab Sci 31(1):3-24.
Abstract: Occupational exposures to inhalation of certain metal dusts or aerosols can cause loss of olfactory acuity, atrophy of the nasal mucosa,

mucosal ulcers, perforated nasal septum, or sinonasal cancer. Anosmia and hyposmia have been observed in workers exposed to Ni- or Cd-containing dusts in alkaline battery factories, nickel refineries, and cadmium industries. Ulcers of the nasal mucosa and perforated nasal septum have been reported in workers exposed to Cr(VI) in chromate production and chrome plating, or to As(III) in arsenic smelters. Atrophy of the olfactory epithelium has been observed in rodents following inhalation of NiSO4 or alpha Ni3S2. Cancers of the nose and nasal sinuses have been reported in workers exposed to Ni compounds in nickel refining, cutlery factories, and alkaline battery manufacture, or to Cr(VI) in chromate production and chrome plating. III animals, several metals (eg, Al, Cd, Co, Hg, Mn, Ni, Zn) have been shown to pass via olfactory receptor neurons from the nasal lumen through the cribriform plate to the olfactory bulb. Some metals (eg. Mn, Ni, Zn) can cross synapses in the olfactory bulb and migrate via secondary olfactory neurons to distant nuclei of the brain. After nasal instillation of a metal-containing solution, transport of the metal via olfactory axons can occur rapidly within hours or a few days (eg, Mn), or slowly other days or weeks (eg, Ni). The olfactory bulb tends to accumulate certain metals (eg, Al, Bi, Cu, Mn, Zn) with greater avidity than other regions of the brain. The molecular mechanisms responsible for metal translocation in olfactory neurons and deposition in the olfactory bulb are unclear, but complexation by metal-binding molecules such as carnosine (beta -alanyl-L-histidine) may be involved.

Song JH, Harris MS, Shin SH. 2001. Effects of fetal bovine serum on ferrous ion-induced oxidative stress in pheochromocytoma (PC12) cells. Neurochem Res 26(4):407-414.
Abstract: Ferrous ion (Fe2+) has been considered to be a cause of neuronal oxidative injury. Since body fluids contain protein and serum is an essential component of tissue culture medium, we have examined the role of serum protein on Fe2+-mediated oxidative stress using PC12 cells and rat cerebral cortices. Fe2+ or the combination of ascorbate and Fe2+ increased concentrations of thiobarbituric acid reactive substances (TBARS) in PC12 cells and cerebrocortical homogenates in medium (RPMI 1640), but did not increase TBARS when the medium was supplemented with 10% fetal bovine serum. Treatment with ascorbate/Fe2+ in serum-free medium reduced endogenous glutathione (GSH) concentration in PC12 cells. However, the medium supplemented with serum did not reduce GSH concentrations. PC12 cell death induced by ascorbate/Fe2+ was alleviated by increasing serum or bovine albumin concentrations in the medium. These observations indicated that oxidative injury caused by the transition metal ion could be lessened by adding fetal bovine serum to culture medium.

Sofic E, Denisova N, Youdim K, Vatrenjak-Velagic V, De Filippo C, Mehmedagic A, Causevic A, Cao G, Joseph JA, Prior RL. 2001. Antioxidant and pro-oxidant capacity of catecholamines and related compounds. Effects of hydrogen peroxide on glutathione and sphingomyelinase activity in pheochromocytoma PC12 cells: potential relevance to age-related diseases. J Neural Transm 108(5):541-557.
Abstract: The antioxidant and pro-oxidant capacity of catecholamines (CA) and related compounds were analyzed using the oxygen radical absorbance capacity (ORAC) assay. In the assay 2,2 ' -azobis (2-amidino-propane) dihydrochloride (AAPH), a peroxyl radical generator, ROOo; H2O2-Cu2+, mainly a hydroxyl radical generator, (OH)-O-o; and Cu2+ a transition metal were used. The antioxidant effect of CA and its related compounds were in the order: neurotransmitters: dopamine (DA), norepinephrine (NE) > metabolites > amino acid precursors as measured by using AAPH. The antioxidant effect of CA and related compounds as measured by using AAPH were linearly correlated with concentration, while the antioxidant effect of CA in scavenging (OH)-O-o produced by H2O2-Cu2 + increased proportionally to concentration at low concentration, but after reaching a maximum declined with increasing concentration. In the presence of Cu2+, CA acted as pro-oxidant. Glutathione (GSH) acted as a pro-oxidant when H2O2-Cu2+ or when CU2+ alone was used as an oxidant

and showed much higher pro-oxidant effect than DA, which could have relevance in the vulnerability of dopaminergic neurons to oxidative stress in the aging and aging related diseases. The antioxidant capacity of CA and many related compounds seems to be correlated with the numbers of hydroxyl groups and their position on the benzoic ring. The 0-methylation and sulfate conjugation of the hydroxyl substitution inactivates both the antioxidant and pro-oxidant activities of CA. Our results show that oxidative stress induced by low (5 muM) or high (300 muM) doses H2O2 in pheochromocytoma PC12 cells significantly up-regulate the activity of Mg-dependent neutral sphingomyelinase (Sase), and significantly decreased GSH.

Serra PA, Rocchitta G, Esposito G, Delogu MR, Migheli R, Miele E, Desole MS, Miele M. 2001. A study on the role of nitric oxide and iron in 3-morpholino-sydnonimine-induced increases in dopamine release in the striatum of freely moving rats. Br J Pharmacol 134(2):275-282.
Abstract: 1 We showed previously that interaction between NO and iron (II), both released following the decomposition of sodium nitroprusside (SNP), accounted for the late SNP-induced dopamine (DA) increase in dialysates from the striatum of freely moving rats; in addition, we showed that coinfusion of iron (II) with the NO-donor S-nitroso-N-acetylpenicillamine mimicked SNP effects on striatal DA release. 2 In the present study, intrastriatal co-infusion of iron (II) (given as FeSO4, 1 mM for 40 min) with the NO-donor and potential peroxynitrite generator 3-morpholinosydnonimine (SIN-1) (0.2, 0.5, 1.0 or 5.0 mM for 180 min), potentiated the SIN-1-induced increase in DA concentration in dialysates from the striatum of freely moving rats. Neither alone nor associated with iron (II) did SIN-1 induce changes in dialysate ascorbic acid or uric acid concentrations. 3 Neither co-infusion of a superoxide dismutase mimetic nor uric acid affected SIN-1-induced increases in dialysate DA concentration. 4 Infusion of the iron chelator deferoxamine (0.2 mm for 180 min) decreased dialysate DA and attenuated SIN-1-induced increases in dialysate DA concentrations. 5 These results suggest that iron plays a key role in SIN-1-induced release of striatal DA and do not support any role for either peroxynitrite or superoxide anion in SIN-1-induced release of striatal DA.

Serra PA, Esposito G, Delogu MR, Migheli R, Rocchitta G, Miele E, Desole MS, Miele M. 2001. Analysis of S-nitroso-N-acetylpenicillamine effects on dopamine release in the striatum of freely moving rats: role of endogenous ascorbic acid and oxidative stress. Br J Pharmacol 132(4):941-949.
Abstract: 1 We showed previously that interaction between NO and iron (II), both released following decomposition of sodium nitroprusside (SNP), accounted for the late SNP-induced dopamine (DA) increase in dialysates from the striatum of freely moving rats. 2 In this study, intrastriatal infusion of the NO-donor S-nitroso-N-acetylpenicillamine (SNAP) (0.2 mM for 180 min) induced a moderate increase in dialysate DA and decreases in ascorbic acid dialysate concentrations; in contrast, SNAP 1 mM infusion induced a long-lasting decrease in both DA and ascorbic acid dialysate concentrations. 3-Methoxy-tyramine (3-MT), dihydroxyphenylacetic acid (DOPAC), homovanillic acid (HVA), and uric acid levels were unaffected. 3 Co-infusion of ferrous sulphate [iron(II), 1 mM for 40 min] with SNAP either 1 or 0.2 mM (for 180 min), produced a significant increase in both DA and 3-MT dialysate concentrations, but it did not affect decreases in dialysate ascorbic acid levels. All other dialysate neurochemicals were unaffected. 4 Co-infusion of ascorbic acid (0.1 mM) with SNAP (1 mM) for 180 min did not modify SNAP-induced decreases in dialysate DA levels. In contrast, co-infusion of uric acid (1 mM) reversed SNAP-induced decreases in dialysate DA, co-infusion of a superoxide dismutase mimetic delayed SNAP-induced DA decreases For a short period, while co-infusion of the antioxidant N-acetylcysteine (NAC, 0.1 mM) significantly increased dialysate DA. 5 The results of this study show that SNAP induces concentration-related changes in DA dialysate levels. At higher concentrations. SNAP induces non-enzymatic DA oxidation, which is inhibited by uric acid and NAG: ascorbic acid failed to protect dialysate DA

from oxidation, probably owing to its promoting effect on SNAP decomposition: exogenous iron(II) may react with NO generated from SNAP decomposition, with a consequent increase in dialysate DA and 3-MT, therefore mimicking SNP effects on striatal DA release.

Schultz C, Dick EJ, Cox AB, Hubbard GB, Braak E, Braak H. 2001. Expression of stress proteins alpha B-crystallin, ubiquitin, and hsp27 in pallido-nigral spheroids of aged rhesus monkeys. Neurobiol Aging 22(4):677-682.
Abstract: Ubiquitin and alpha B-crystallin belong to a class of proteins which are overexpressed in a variety of human neuropathological conditions associated with increased cellular stress. In this study we have examined the brains of aged rhesus monkeys (Macaca mulatta; n = 10, mean age: 39.7 years) using antibodies against the stress proteins ubiquitin, alpha B-crystallin. and heat shock protein 27 (hsp27). Here, we demonstrate an increased expression of ubiquitin, alpha B-crystallin, and hsp27 in spheroid bodies predominantly localized in the globus pallidus and pars reticulata of the substantia nigra. A portion of the pallido-nigral spheroids also contained ferric iron as highlighted by Perls' staining. On the basis of these findings we advance the hypothesis that expression of ubiquitin, alpha B-crystallin, and hsp27 in pallido-nigral spheroids of aged rhesus monkeys represents a stress response possibly related to increased iron-mediated oxidative stress. (C) 2001 Elsevier Science Inc. All rights reserved.

Schroder N, Fredriksson A, Vianna MRM, Roesler R, Izquierdo I, Archer T. 2001. Memory deficits in adult rats following postnatal iron administration. Behav Brain Res 124(1):77-85.
Abstract: Two experiments investigated the effects of Fe2+, administered postnatally to rat pups on days 10-12, upon tests of memory performance and motor behaviour. In experiment I, Wistar rat pups were administered Fe2+ at doses of either 2.5, 7.5, 15.0 or 30.0 mg/kg, or vehicle, postnatally, and tested in the open-field at 3 months of age, followed 6 weeks later by testing in the radial arm maze. In the open-field test, only the 30.0 mg/kg Fe2+ group showed a significantly decreased number of ambulations, but not rearings. In the radial arm maze, all four dose groups, demonstrated deficits in acquisition performance from test days 3 to 5. Retention quotients confirmed the cognitive deficits over all four Fe2+ groups. In experiment II, rats were administered either 2.5, 7.5 or 22.5 mg Fe2+ per kg, or vehicle, postnatally, and tested in the inhibitory avoidance (IA) conditioning and retention test at 3 months of age. In the IA conditioning test, groups were either given five 10-min preexposures to the test chamber (preexposed) or simply moved to another cage (non-preexposed). IA retention was blocked in non-preexposed rats administered 7.5 and 22.5 mg Fe2+ per kg whereas in preexposed rats the 7.5 mg/kg group did not differ from the control (vehicle) group, although the preexposed control group showed significantly better retention than the non-preexposed control group. Postnatal iron administration appears to induce long-lasting detrimental effects upon performance of both appetitively and negatively reinforced tests of memory. Analysis of iron content indicated significant increases in the substantia nigra of the 7.5, 15.0 and 30.0 mg/kg dose groups, but not in the 2.5 mg/kg dose group. Postnatal iron administration appears to induce far-reaching effects upon the performance of certain learned behaviours, (C) 2001 Elsevier Science B.V. All rights reserved.

Sayre LM, Smith MA, Perry G. 2001. Chemistry and biochemistry of oxidative stress in neurodegenerative disease. Curr Med Chem 8(7):721-738.
Abstract: The age-related neurodegenerative diseases exemplified by Alzheimer's disease (AD), Lewy body diseases such as Parkinson's disease (PD), amyotrophic lateral sclerosis (ALS), and Huntington's disease are characterized by the deposition of abnormal forms of specific proteins in the brain. Although several Factors appear to underlie the pathological depositions, the cause of neuronal death in each disease appears to be multifactorial. In this regard, evidence in each case for a role of oxidative stress is provided by the finding that the pathological deposits are

immunoreactive to antibodies recognizing protein side-chains modified either directly by reactive oxygen or nitrogen species, or by products of lipid peroxidation or glycoxidation. Although the source(s) of increased oxidative damage are not entirely clear, the findings of increased localization of redox-active transition metals in the brain regions most affected is consistent with their contribution to oxidative stress. It is tempting to speculate that free radical oxygen chemistry plays a pathogenetic role in all these neurodegenerative conditions, though it is as yet undetermined what types of oxidative damage occur early in pathogenesis, and what types are secondary manifestations of dying neurons. Delineation of the profile of oxidative damage in each disease will provide clues to how the specific neuronal populations are differentially affected by the individual disease conditions.

Ren MQ, Xie JP, Wang XS, Ong WY, Leong SK, Watt F. 2001. Iron concentrations and distributions in the parkinsonian substantia nigra of aged and young primate models. Nuclear Instruments & Methods in Physics Research Section B-Beam Interactions With Materials and Atoms 181:522-528.
Abstract: Parkinson's disease (PD) is a progressive neuronal degenerative brain disease of the elderly, and is caused by the selective degeneration of neurons in the substantia nigra (SN) region of the brain, resulting in a reduced production of the neurotransmitter dopamine. Iron has been linked to dopaminergic cell death in Parkinson's disease because of its potential to promote free radicals, leading to oxidative stress. The present study is aimed at using the techniques of nuclear microscopy to elucidate the iron concentrations and distributions in the SN of both young and old monkeys following unilateral 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP)-lesioning. A group of three old monkeys (older than 7 years) and a group of three young monkeys (younger than 7 years) were unilaterally MPTP-lesioned (right side) to induce parkinsonism and sacrificed after 35 days. The left side SN was used as a control. This time interval was chosen to correspond to an average 50% loss of dopamine producing cells in the lesioned right side SN, We have observed a significant difference in iron concentrations between the SNs of the young and old monkeys (increasing from an average of 233 to 1092 parts per million dry weight). When comparing the lesioned and non-lesioned SNs of the same animal, we found no significant difference in iron levels for each young monkey. However we have found a slight increase in iron (approximately 10%) between the lesioned SN and control SN for old monkeys. We have also observed that in the SN of younger primates, there is a weak anti-correlation in the SN iron levels with the neuron distribution. In the older monkeys, however, we have observed a proliferation of iron-rich granules, which appear to be more strongly anti-correlated with the distribution of neurons. The iron-cell anti-correlation occurs both in the control as well as the lesioned SN. Our results suggest that iron, particularly in the form of iron-rich deposits, accumulates in specific sites in the SN with age. Since Parkinson's disease mainly occurs in the elderly, this may implicate iron as a factor in dopaminergic cell death through iron-catalysed free radical production. (C) 2001 Elsevier Science B.V. All rights reserved.

Rein FN, Rocha RC, Toma HE. 2001. Catecholamine complexes of ruthenium-edta and their redox chemistry. J Inorg Biochem 85(2-3):155-166.
Abstract: The electrochemical and spectroelectrochemical behavior of some neurotransmitters (dopamine and L-dopa) and their corresponding novel blue ruthenium(III)-edta complexes were investigated in aqueous solutions. At pH 7-10, the free ligand species can be electrochemically oxidized in the range of 0.1-0.6 V versus SHE, yielding primarily quinone products susceptible to pH-dependent, secondary intramolecular chemical reactions, which make the redox processes irreversible. When coordinated to the ruthenium(III)-edta complex, their electrochemical and spectroelectrochemical behavior is dramatically changed, approaching that of metal complexes with noninnocent dioxolene ligands. Reduction of the ruthenium(III) moiety proceeds reversibly above pH 9, in the region from -0.5 to -0.7 V. The oxidation process centered on the catecholate ligands becomes reversible and leads exclusively to thr formation of the

semiquinone species, with no evidence of complications from further reactions. These changes in the electrochemical behavior of the neurotransmitters make their cyclovoltammetric waves for reduction/ oxidation more defined, favoring more precise quantitative analyses. (C) 2001 Elsevier Science B.V. All rights reserved.

Reichmann H, Summer U, Gerlach M, Riederer P . 2001. Pharmacotherapy of idiopathic Parkinson's syndrome with special focus on neuroprotection. Nervenheilkunde 20(4):227-+.
Abstract: Most of the commonly used antiparkinsonian drugs show neuroprotective potency when tested in tissue culture or animal models. Neuroprotection consists of measures which lead to prevention or delay of neuronal cell death. So far, there are no clinical studies which show undoubtably neuroprotection. Nonetheless, there ore 3 PET- or SPECT-controlled studies with ropinirole, pergolide and pramipexole finished which were designed to prove neuroprotection while taking dopamine agonists. This paper will further introduce studies with selegiline and NMDA receptor antagonists which indicate possible neuroprotection. Experimental data suggest studies with radical scavengers, coenzyme Q, iron chelators or antiopoptotic drugs such as flupirtine. Taking all consisting data into account we recommend to treat early Parkinsonism with a combination of selegiline, NMDA receptor antagonists and dopamine agonists.

Ravaglia G, Forti P, Maioli F, Bianchi GP, Vettori C, Nesi B, Sacchetti L, Orlanducci P, Cavalli G. 2001. Oxidative stress and dementia in oldest-old subjects. Arch Gerontol Geriatr :325-331.
Abstract: The possible involvement of oxidative damage and antioxidant protection has been suggested in the pathogenesis of dementia, a cognitive disorder whose prevalence increase with aging. In this study we investigated the relationship between dementia and plasma status of antioxidants and oxidative products in oldest-old subjects. Plasma levels of thiobarbituric acid-reactive substances (TBARS), vitamin A, vitamin E, coenzyme Q(10) (CoQ(10)), and serum levels of selenium (Se) and zinc (Zn) were determined in 30 demented patients aged 90 to 107 years in comparison with 32 normal individuals aged 90 to 103 years. Dementia was diagnosed according to DSM-IV criteria. TBARS, vitamin A, vitamin E and CoQ(10) were measured by HPLC method. Zn and Se were measured by absorption spectrometry. Vitamins were expressed as lipid-adjusted concentrations. Demented men had higher plasma TBARS ($p = 0.037$) and lower plasma lipid-adjusted vitamin E ($p = 0.043$) than normal men, whereas no difference was found for the remaining variables. No difference was found between demented and normal women for the variables of interest. In oldest-old men, but not in women, dementia seems to be associated with an enhanced lipid peroxidation and a reduced vitamin E reserve. It remains to be resolved, however, as to whether this enhanced oxidative stress is a cause or a result of lowered antioxidants.

Racette BA, Perry A, D'avossa G, Perlmutter JS. 2001. Late-onset neurodegeneration with brain iron accumulation type 1: Expanding the clinical spectrum. Mov Disord 16(6):1148-1152.
Abstract: We report on two patients with pathologically proven neurodegeneration with brain iron accumulation type 1 (NBIA-1) with late onset and atypical presentations. One patient experienced gradual onset of shuffling gait, rigidity, bradykinesia, and increasing postural instability at age 85 years. He died a few weeks after developing acute hemiballismus at age 90 years. Histopathology revealed marked neuronal loss in the internal segment of the globus pallidum, astrocytosis, axonal spheroids, and extensive iron deposition consistent with NBIA-1. No additional lesions were found to explain the hemiballismus. The second patient experienced fulminant dementia evolving to total disability and death within 2 months. Autopsy showed typical NABIA-1 pathology. We conclude that NBIA-1 pathology can develop at any age, and that the phenotype should be expanded to include late-onset parkinsonism. The relationship to hemiballismus and adult-onset dementia is less clear. (C) 2001 Movement

Disorder Society.

Ponting CP. 2001. Domain homologues of dopamine beta-hydroxylase and ferric reductase: roles for iron metabolism in neurodegenerative disorders? Hum Mol Genet 10(17):1853-1858.
Abstract: One of the defining characteristics of neurodegenerative diseases, including Parkinson's, Alzheimer's and Huntington's diseases, is abnormal accumulations of iron, specifically in affected areas. Following injection of iron in rat brains, a relatively selective lesion of dopamine neurons, similar to parkinsonism, occurs. These observations indicate that Fe(II)-mediated generation of free radical species, by the Fenton reaction, might contribute to the pathoetiology of these diseases. Iron is known to possess multiple roles in the biosynthesis of catecholamines in dopaminergic neurons. These include, as Fe(II), facilitating the production of dopamine from phenylalanine by tyrosine hydroxylase, and as heme, assisting the recycling of ascorbate by cytochrome b-561 required for the generation of norepinephrine from dopamine by dopamine beta - hydroxylase. In this study, it is demonstrated that a human and mouse gene product, stromal cell-derived receptor 2, is a homologue of cytochrome b-561 and duodenal cytochrome b, and is thus predicted to be active as a ferric reductase. Moreover, this protein also contains a domain homologous to the N-terminal regulatory region of dopamine beta - hydroxylase. These findings from sequence analysis lead to a prediction that stromal cell-derived receptor 2 is a catecholamine-regulated ferric reductase active in the brain. Dysfunction of cytochrome b-561 or stromal cell-derived receptor 2, therefore, might predispose individuals to abnormal accumulation of Fe(III) and/or generation of cytotoxic free radicals as a consequence of a rapid cycling between Fe(III) and Fe(II). The hypothesis that aberrant ferric reductase activities are involved in the progression of neurodegenerative diseases should open up new avenues of research, and possibly therapy, for these devastating diseases.

Pinero DJ, Jones BC, Beard JL. 2001. Variations in dietary iron alter behavior in developing rats. J Nutr 131(2):311-318.
Abstract: Iron deficiency in children is associated with retardation in growth and cognitive development, and the effects on cognition may be irreversible, even with treatment. Excessive iron has also been associated with neurological disease, especially in reference to the increased iron content in the brains of Alzheimer's disease and Parkinson's disease patients. This study evaluated the effects of dietary iron deficiency and excess iron on physical activity in rats. The animal model used is developmentally sensitive and permits control of the timing as well as the duration of the nutritional insult. Hence, to study the effects of early, late and long-term iron deficiency or excess iron (supplementation), rats were either made iron deficient or supplemented on postnatal day (PND) 10-21, PND 21-35 and PND 10-35. Some iron-deficient rats were iron repleted between PND 21-35. Different measures of motor activity were taken at PND 14, 17, 20, 27 and 34. Iron-deficient and iron-supplemented rats showed decreased activity and stereotypic behavior; this was apparent for any onset and duration of the nutritional insult. Recovery from iron deficiency did not normalize these functional variables, showing that the deleterious effects of early iron deficiency persist despite subsequent adequate treatment. This study demonstrates that iron deficiency in early life leads to irreversible behavioral changes. The biological bases for these behavioral alterations are not readily apparent, because iron therapy rapidly reverses the iron losses in all brain regions.

Pifl C, Zezula R, Spittler A, Kattinger A, Reither H, Caron MG, Hornykiewicz O. 2001. Antiproliferative action of dopamine and norepinephrine in neuroblastoma cells expressing the human dopamine transporter. FASEB J 15(7).
Abstract: The classical catecholamine transmitters dopamine and norepinephrine are also involved in neurodevelopment and neuron cell death. These effects are thought to be mediated by receptors coupled to intracellular second messengers and by oxidative stress, respectively.

Using human SK-N-MC neuroblastoma cells stably transfected with the dopamine or norepinephrine transporter, we show that catecholamines have profound effects on cell viability not mediated by any of the above mechanisms. Low micromolar concentrations of dopamine or norepinephrine (1-10 muM) affected cell growth depending on the expression level of plasmalemmal transport. The growth inhibition was accompanied by a profound effect on the cell cycle profile with an arrest in G, as determined by flow cytometry measuring DNA content. In a proportion of cells, apoptosis was present. Various antioxidants did not prevent the catecholamine effect, and there was no indication of accumulation of reactive oxygen species (as determined by hydroethidine, dichlorofluorescein, and the inactivation-reactivation pattern of aconitase), ruling out involvement of oxidative stress. Reversal by FeCl3 and parallel effects of the iron-chelator deferoxamine suggest that the catecholamine action was related to intracellular iron chelation. This novel biological action of dopamine and norepinephrine unrelated to neurotransmitter receptors, second messengers, or oxidative stress may be important for cell differentiation during neurodevelopment and survival of differentiated neurons.

Piemonte F, Pastore A, Tozzi G, Tagliacozzi D, Santorelli FM, Carrozzo R, Casali C, Damiano M, Federici G, Bertini E. 2001. Glutathione in blood of patients with Friedreich's ataxia. Eur J Clin Invest 31(11):1007-1011.
Abstract: Background Oxidative stress and mitochondrial dysfunction have long been considered to play a role in Friedreich's ataxia, a neurodegenerative disease due to a GAA expansion in a gene coding for a mitochondrial protein (frataxin), implicated in the regulation of iron metabolism. Since glutathione is an important antioxidant whose role has been recently proposed in the pathogenesis of some neurodegenerative diseases, we investigated glutathione metabolism in the blood of 14 patients with Friedreich's ataxia by measuring total, free and protein-bound glutathione concentrations. Materials and methods Blood samples were obtained from 14 unrelated patients with Friedreich's ataxia (nine males, five females) and 20 age-matched healthy controls (10 males, 10 females). Total and free glutathione concentrations were determined by reverse-phase liquid chromatography with fluorescence detection; the glutathionyl-haemoglobin separation from healthy and pathological subjects was obtained by electrospray ionization-mass spectrometry. Results We consistently found a reduction of free glutathione levels (0.55 +/- 0.06 nmol mg(-1) haemoglobin, vs. 8.4 +/- 1.79 nmol mg(-1) haemoglobin, P < 0.001) in the blood of patients with Friedreich's ataxia, a total glutathione concentration comparable to the controls (15 +/- 2-6 nmol mg(-1) haemoglobin, vs. 15.4 +/- 1.4 nmol mg(-1) haemoglobin), and a significant increase of glutathione bound to haemoglobin (15 +/- 1.5 vs. 8 +/- 1.8%, P < 0.05) in erythrocytes. Conclusions Our findings give evidence of an impairment in vivo of glutathione homeostasis in Friedreich's ataxia, suggesting a relevant role of free radical cytotoxicity in the pathophysiology of the disease; this study may also prove useful in the search for an oxidative stress marker in neurodegeneration.

Piemonte F, Casali C, Carrozzo R, Schagger H, Patrono C, Tessa A, Tozzi G, Cricchi F, Di Capua M, Siciliano G, Amabile GA, Morocutti C, Bertini E, Santorelli FM. 2001 . Respiratory chain defects in hereditary spastic paraplegias. Neuromuscul Disord 11(6-7):565-569.
Abstract: Hereditary Spastic Paraplegias (HSPs) are heterogeneous neurodegenerative disorders whose etiopathogenesis is still unclear. The identification of pathogenic mutations in a gene (SPG7) encoding a mitochondrial metalloprotease suggested that oxidative phosphorylation (OXPHOS) alterations might underlie HSP in a subgroup of patients. We performed clinical, morphological, biochemical, and molecular genetic studies in six HSP patients and in six sporadic patients to investigate OXPHOS in muscle biopsies. Complicated and pure forms were included in our study. Morphological alterations of the type seen in OXPHOS-related disorders were found in three patients. Five patients showed an isolated defect of complex I activity. No mutations in the SPG7 gene were detected.

Our results suggest that OXPHOS defects in HSP patients are more common than previously believed. (C) 2001 Elsevier Science B.V. All rights reserved.

Petkova V. 2001. [Diagnostic problems of post-intoxication states]. Med Tr Prom Ekol (4):21-4.
Abstract: Complete clinical examination covered 37 patients in various stages after acute occupational poisonings with pesticides (8 examinees), with explosive gases (9 subjects), with carbon oxide (3 examinees), with cadmium oxides (1 examinee), with nitrogen compounds (4 ones), with sulfurous gases (2 examinees), with organic solvents (10 subjects) including chlorinated hydrocarbons (2 subjects). Post-intoxication period appeared to include syndromes characteristic for each poison and corresponding to severity of acute state (from cephalgia to parkinsonism and thyrotoxicosis after acute poisoning with carbon oxide, organic neurologic signs and toxic hepatitis after acute poisoning with phosphorus organic compounds, cerebral asthenia after acute exposure to organic solvents, severe encephalopathy and toxic auditory and optic neuropathy after hydrogen sulfide). Inadequate medical rehabilitation and continuous occupational exposure induced deterioration in clinical signs and advanced post-intoxication syndromes.

Orth M, Schapira AHV. 2001. Mitochondria and degenerative disorders. Am J Med Genet 106(1):27-36.
Abstract: In mammalian cells, mitochondria provide energy from aerobic metabolism. They play an important regulatory role in apoptosis, produce and detoxify free radicals, and serve as a cellular calcium buffer. Neurodegenerative disorders involving mitochondria can be divided into those caused by oxidative phosphorylation (OXPHOS) abnormalities either due to mitochondrial DNA (mtDNA) abnormalities, e.g., chronic external ophthalmoplegia, or due to nuclear mutations of OXPHOS proteins, e.g., complex I and II associated with Leigh syndrome. There are diseases caused by nuclear genes encoding non-OXPHOS mitochondrial proteins, such as frataxin in Friedreich ataxia (which is likely to play an important role in mitochondrial-cytosolic iron cycling), paraplegin (possibly a mitochondrial ATP-dependent zinc metalloprotease of the AAA-ATPases in hereditary spastic paraparesis), and possibly Wilson disease protein tan abnormal copper transporting ATP-dependent P-type ATPase associated with Wilson disease). Huntingon disease is an example of diseases with OXPHOS defects associated with mutations of nuclear genes encoding non-mitochondrial proteins such as huntingtin. There are also disorders with evidence of mitochondrial involvement that cannot as yet be assigned. These include Parkinson disease (where a complex I defect is described and free radicals are generated from dopamine metabolism), amyotrophic lateral sclerosis, and Alzheimer disease, where there is evidence to suggest mitochondrial involvement perhaps secondary to other abnormalities. (C) 2001 Wiley-Liss, Inc.

Obata T, Yamanaka Y. 2001. Nitric oxide enhances MPP+-induced hydroxyl radical generation via depolarization activated nitric oxide synthase in rat striatum. Brain Res 902(2):223-228.
Abstract: We examined the effect of NG-nitro-L-arginine methyl ester (L-NAME), a nitric oxide synthase (NOS) inhibitor, on extracellular potassium ion concentration ([K+](o))-enhanced hydroxyl radical (OH) generation due to 1-methyl-4-phenylpyridinium ion (MPP+) was examined in the rat striatum. Rats were anesthetized, and sodium salicylate in Ringer's solution (0.5 nmol/mul per min) was infused through a microdialysis probe to detect the generation of OH as reflected by the non-enzymatic formation of 2,3-dihydroxybenzoic acid (DHBA) in the striatum. Induction of KCl (20, 70 and 140 mM) increased MPP+-induced OH formation trapped as 2,3-dihydroxybenzoic acid (DHBA) in a concentration dependent manner. However, the application of L-NAME (5 mg/kg i.v.) abolished the [K+], depolarization-induced OH formation with MPP+. Dopamine (DA; 10 muM) also increased the levels of DHBA due to MPP+. However, the effect of DA after application of L-NAME did not change the levels of DHBA. On the other

hand, the application of allopurinol (20 mg/kg i.v., 30 min prior to study), a xanthine oxidase (XO) inhibitor was abolished the both [K+](o)- and DA-induced OH generation. Moreover, when iron(II) was administered to MPP+ then [K+](o) (70 mM)-pretreated animals, a marked increase in the level of DHBA. However, when corresponding experiments were performed with L-NAME-pretreated animals, the same results were obtained. Therefore, NOS activation may be no relation to Fenton-type reaction via [K+](o) depolarization-induced OH generation. The present results suggest that [K +](o)-induced depolarization augmented MPP+-induced OH formation by enhancing NO synthesis. (C) 2001 Elsevier Science B.V. All rights reserved.

Nappi AJ, Vass E. 2001. The effects of nitric oxide on the oxidations of L-dopa and dopamine mediated by tyrosinase and peroxidase. J Biol Chem 276 (14):11214-11222.
Abstract: The effects of nitric oxide (NO) on both tyrosinase/O-2- and horseradish peroxidase/H2O2-mediated oxidations of dopamine and its o-dihydric phenol precursor L-dopa were compared with autoxidative processes and quantitatively assessed by oxidative and reductive electrochemical detection systems. In peroxidase/H2O2/NO-catalyzed reactions, significantly more substrate was oxidized than in the corresponding control incubations lacking NO. In tyrosinase/O-2/NO-promoted reactions the total amounts of L-dopa and dopamine oxidized were significantly less than the amounts of the substrates oxidized by enzyme alone. These data indicate that the activity of the heme protein peroxidase was enhanced by NO, whereas tyrosinase, a copper-containing monoxygenase, was inhibited. The NO-mediated reduction of tyrosinase/ O-2 activity may be attributed to the formation of an inhibitory copper nitrosyl complex. An oxidized nitrodopamine derivative, considered to be either the quinone or semiquinone of 6-nitrosodopamine, was generated in peroxidase/H2O2/NO-mediated reactions with dopamine along with two oxidized melanin precursors, dopamine quinone and dopaminechrome. No corresponding nitroso compound was formed in reactions involving L-dopa or in any of the tyrosinase-mediated reactions, The formation of such a noncyclized nitrosodopamine represents an important alternative pathway in catecholamine metabolism, one that by-passes the formation of cytoprotective indole precursors of melanin. The results of this investigation suggest that cellular integrity and function can be adversely affected by NO-promoted oxidations of dopamine and other catechols, reactions that not only accelerate their conversion to reactive quinones but also form potentially cytotoxic noncyclized nitroso derivatives. Reduced levels of dopamine in the brain through NO-enhanced oxidation of the catecholamine will almost certainly be manifested by diminished levels of the dopamine-derived brain pigment neuromelanin.

Napolitano A, Di Donato P, Prota G. 2001. Zinc-catalyzed oxidation of 5-S-cysteinyldopa to 2,2 '-bi(2H-1,4-benzothiazine): Tracking the biosynthetic pathway of trichochromes, the characteristic pigments of red hair. J Org Chem 66(21):6958-6966.
Abstract: Trichochromes, the peculiar pigments of red human hair, featuring the Delta (2,2')-bi(2H-1,4-benzothiazine) skeleton, are known to arise from cysteinyldopas, mainly the 5-S-isomer (5). However, the mode of formation and the direct precursors have remained largely undefined. To fill this gap, we investigated the oxidation of 5 in air or with chemical and enzymatic agents under biomimetic conditions. In the presence of zinc ions, which occur in epidermal tissues at significant concentrations, the reaction course is diverted toward the formation of a labile 3-carboxy-2H-1,4-benzothiazine intermediate (11), which was identified by direct NMR analysis. Structural formulation was supported by characterization of the analogous compound 13 isolated from oxidation of the model 5-methyl-3-S-cysteinylcatechol (12) after methylation. In the further stages of the oxidation, diastereomeric 2,2'-bi(2H-1,4-benzothiazine) 15 and 14 were obtained from 5 and 12, respectively, the reaction proceeding at a higher rate and to a greater extent in the presence of acids. The dimers were shown to readily convert to each other in the presence of acids. In the case of the methylated dimers 14, a 2,2'-bi

(4H-1,4-benzothiazine) intermediate (16) was isolated and characterized. In acidic media, trichochrome C (1a), the most abundant in red human hair, was smoothly formed from aerial oxidation of 15, and under similar conditions, trichochrome-related products (17 and 18) were obtained from 14 prior to or after methylation. The presence of la and precursors 5 and 15 was investigated by HPLC analysis of red hair samples following mild proteolytic digestion. On the basis of these data, a likely biosynthetic route to trichochrome pigments of red human hair is depicted.

Mwanjewe J, Hui BK, Coughlin MD, Grover AK. 2001. Treatment of PC12 cells with nerve growth factor increases iron uptake. Biochem J 357:881-886.
Abstract: Phaeochromocytoma PC12 cells treated with nerve growth factor (NGF) differentiate into a neuronal phenotype. Here we compare the uptake of transferrin-bound and non-transferrin-bound iron in NGF-treated (neuronal phenotype) and control (proliferating) PC12 cells. The non-transferrin-bound iron uptake was greater in the NGF-treated cells than in the control, independently of the uptake time, the iron-chelating agents used, the oxidation state of iron (Fe2+ or Fe2+) and the iron concentration tested. The NGF-treated cells expressed L-type and N-type voltage-operated Ca2+ channels. Nitrendipine (an L-type inhibitor) and possibly omega -conotoxin (an N-type inhibitor) inhibited the iron uptake by 20 %. Thapsigargin inhibits the endoplasmic reticulum Ca2+ pump and allowed Mn2+ entry into cells. Preincubating PC12 cells with thapsigargin increased the iron uptake. The rate of transferrin-bound iron uptake was less than 1 % of the non-transferrin-bound iron uptake and the maximum transferrin-bound iron uptake was also very low. We conclude that an increase in the iron uptake by multiple pathways accompanies the transition of PC12 cells from the proliferating to the neuronal phenotype.

Momcilovic B, Alkhatib HA, Duerre JA, Cooley M, Long WM, Harris TR, Lykken GI. 2001. Environmental lead-210 and bismuth-210 accrue selectively in the brain proteins in Alzheimer disease and brain lipids in Parkinson disease. Alzheimer Disease & Associated Disorders 15(2):106-115.
Abstract: We studied the occurrence of the environmental radon daughters, Po-210 (alpha particles), and Bi-210 (beta particles), in the protein and lipid fractions of cortical gray and subcortical white matter from the frontal and temporal lobes of human brains of persons with Alzheimer disease (AD), persons with Parkinson disease (PD), smokers, or persons with no previous evidence of clinical neurologic disease (controls). We found a 10-fold increase in Po-210 and Pb-210 radioactivity in the protein fraction from both the cortical gray and subcortical white matter in AD and smokers, and a similar increase in the lipid fraction in PD. The pathognomonic distribution of the radon daughters to the lipids in PD and to the proteins in AD was inferred to reflect the increase of local chlorine availability to which radon daughters bound selectively. Cigarette smoking strongly increases radon daughter retention in the central nervous system.

Milanese M, Ibn Lkhayat M, Zatta P. 2001. Inhibitory effect of aluminum on dopamine beta-hydroxylase from bovine adrenal gland. J Trace Elem Med Biol 15(2-3):139-141.
Abstract: Aluminum is a well known neurotoxic agent that is overaccumulated in the substantia nigra of patients affected by Parkinson's disease as well as in certain cerebral areas of other neurodegenerative pathologies such as Alzheimer's disease. Although the role of aluminum in neurodegenerative diseases is yet to be clearly understood, the metal ion is known to substantially alter the activity of several key enzymes in the central nervous system. The present paper reports data on the effect of aluminum on the activity of dopamine-beta-hydroxylase from bovine adrenal gland utilized as a model study. The metal ion inhibited the activity of this enzyme with a mixed type mechanism following the Michaelis-Menten equation. In the absence of Al, the enzyme exhibited a Km and Vmax of 2.56 mM of 4.12 pmol/min respectively, while in the presence of Al its Km and Vmax were 3.85 mM and 2.86 pmol/min respectively. The potential implications of aluminum in the etiopathogenesis of neurological

disorders are discussed.

Mendez-Ivarez E, Soto-Otero R, Hermida-Ameijeiras A. 2001. Effects of aluminium and zinc on hydroxyl free radical generation during 6-hydroxydopamine autoxidation: relevance for Parkinson's disease. J Neurochem 77:42.

Mendez-Alvarez E, Soto-Otero R, Hermida-Ameijeiras A, Lopez-Martin ME, Labandeira-Garcia JL. 2001. Effect of iron and manganese on hydroxyl radical production by 6-hydroxydopamine: Mediation of antioxidants . Free Radic Biol Med 31(8): 986-998.
Abstract: 6-Hydroxydopamine (6-OHDA) neurotoxicity has often been related to the generation of free radicals. Here we examined the effect of the presence of iron (Fe2+ and Fe3+) and manganese and the mediation of ascorbate, L-cysteine (CySH), glutathione (GSH), and N-acetyl-CySH on hydroxyl radical ((OH)-O-.) production during 6-OHDA autoxidation. In vitro, the presence of 800 nM iron increased (> 100%) the production of (OH)-O-. by 5 muM 6-OHDA while Mn2+ caused a significant reduction (72%). The presence of ascorbate (100 muM) induced a continuous generation of (OH)-O-. while the presence of sulfhydryl reductants (100 muM) limited this production to the first minutes of the reaction. In general, the combined action of metal + antioxidant increased the (OH)-O-. production, this effect being particularly significant (> 400%) with iron + ascorbate. In vivo, tyrosine hydroxylase immunohistochemistry revealed that intrastriatal injections of rats with 6-OHDA (30 nmol) + ascorbate (600 nmol), 6-OHDA + ascorbate + Fe2+ (5 nmol), and 6-OHDA + ascorbate + Mn2+ (5 nmol) caused large striatal lesions, which were markedly reduced (60%) by the substitution of ascorbate by CySH. Injections of Fe2+ or Mn2 + alone showed no significant difference to those of saline. These results clearly demonstrate the role of ascorbate as an essential element for the neurotoxicity of 6-OHDA, as well as the diminishing action of sulfhydryl reductants, and the negliglible effect of iron and manganese on 6-OHDA neurotoxicity. (C) 2001 Elsevier Science Inc.

Martinez A, Knappskog PM, Haavik J. 2001. A structural approach into human tryptophan hydroxylase and its implications for the regulation of serotonin biosynthesis. Curr Med Chem 8(9):1077-1091.
Abstract: Tryptophan hydroxylase (TPH) catalyzes the 5-hydroxylation of tryptophan, which is the first step in the biosynthesis of indoleamines (serotonin and melatonin). Serotonin functions mainly as a neurotransmitter, whereas melatonin is the principal hormone secreted by the pineal gland. TPH belongs to the family of the aromatic amino acid hydroxylases, including phenylalanine hydroxylase (PAH) and tyrosine hydroxylase (TH), which all have a strict requirement for dioxygen, non-heme iron (II) and tetrahydrobiopterin (BH4). During the last three years there has been a formidable increase in the amount of structural information about PAH and TH, which has provided new insights into the active site structure, the binding of substrates, inhibitors and pterins, as well as on the effect of disease-causing mutations in these hydroxylases. Although structural information about TPH is not yet available, the high sequence homology between the three mammalian hydroxylases, notably at the catalytic domains, and the similarity of the reactions that they catalyze, indicate that they share a similar 3D-structure and a common catalytic mechanism. Thus, we have prepared a model of the structure of TPH based on the crystal structures of TH and PAH. This structural model provides a frame for understanding the specific interactions of TPH with L-tryptophan and substrate analogues, BH4 and cofactor analogues, L-DOPA and catecholamines. The interactions of these ligands with the enzyme are discussed focusing on the physiological and pharmacological regulation of serotonin biosynthesis, notably by tryptophan supplementation therapy and substitution therapy with tetrahydrobiopterin analogues positive effects), as well as the effect of catecholamines on TPH activity in L-DOPA treated Parkinson's disease patients (enzyme inhibition).

Martin WR. 2001. Magnetic resonance imaging and spectroscopy in Parkinson's

disease. Adv Neurol 86:197-203.
Abstract: At present, conventional MR imaging shows no convincing structural changes in PD itself but may be useful in helping to distinguish PD from other neurodegenerative parkinsonian syndromes and from the occasional case of parkinsonism secondary to a focal brain lesion. MR spectroscopy may also provide useful information in distinguishing PD from disorders such as MSA. The general field of MR imaging and spectroscopy is evolving rapidly, and there are a number of areas in which we can expect new developments to provide relevant information. Novel pulse sequences may provide more information regarding substantia nigra pathology in PD. The use of MR as a tool to measure regional iron concentrations should provide more information regarding the relationship between iron accumulation and parkinsonian symptoms. MR spectroscopy provides a sensitive tool for the researcher to investigate in vivo the possible contribution of abnormalities in brain energy metabolism to the pathogenesis of PD. Spectroscopy also allows the assessment of other metabolite changes in PD, for example, providing for the evaluation of the potential importance of changes in regional brain glutamate content. Last, although not considered in the present review, functional MR imaging provides the potential to evaluate, in a noninvasive fashion, the role played by the basal ganglia in motor control and in cognition in normal individuals as well as in PD.

Manini P, D'ischia M, Prota G. 2001. An unusual decarboxylative Maillard reaction between L-DOPA and D-glucose under biomimetic conditions: Factors governing competition with Pictet-Spengler condensation. J Org Chem 66 (15):5048-5053.
Abstract: In 0.1 M phosphate buffer at pH 7.4 and 37 degreesC, the tyrosine metabolite L-3,4-dihydroxyphenylalanine (L-DOPA) reacts smoothly with D-glucose to afford, besides diastereoisomeric tetrahydroisoquinolines 1 and 2 by Pictet-Spengler condensation, a main product shown to be the unexpected decarboxylated Amadori compound N-(1-deoxy-D-fructos-1-yl)-dopamine (3). Under similar conditions, dopamine gave only tetrahydroisoquinoline products 4 and 5, whereas L-tyrosine gave exclusively the typical Amadori compound 6. Fe3+ and Cu2+ ions, which accumulate in relatively high levels in parkinsonian substantia nigra, both inhibited the formation of 3. Cu2+ ions also inhibited the formation of I and 2 to a similar degree, whereas Fe3+ ions increased the yields of I and 2. Apparently, the formation of 3 would not be compatible with a simple decarboxylation of the initial Schiff base adduct, but would rather involve the decarboxylative decomposition of a putative oxazolidine-5-one intermediate assisted by the catechol ring. These results report the first decarboxylative Maillard reaction between an amino acid and a carbohydrate under biomimetic conditions and highlight the critical role of transition metal ions in the competition with Pictet-Spengler condensation.

Manini P, D'ischia M, Prota G. 2001. Pictet-Spengler condensation of the antiparkinsonian drug L-DOPA with D-glyceraldehyde. Opposite kinetic effects of Fe3+ and Cu2+ ions and possible implications for the origin of therapeutic side effects. Bioorganic & Medicinal Chemistry 9(4):923-929.
Abstract: In 0.05 M phosphate buffer, pH 7.4. and at 37 degreesC, L-DOPA. a widely used antiparkinsonian drug, reacted smoothly with D-glyceraldehyde to afford diastereoisomeric (1R,1 'S,3S)-3-carboxy-1-(1 ' . 2 ' -dihydroxyethyl)-6,7-dihydroxy- 1,1,3,4-tetrahydroisoquinoline (1) and (1S,1 'S,3S)-3-carboxy-1-(1 ' ,2 ' -dihydroxyethyl)-6,7-dihydroxy-1,2,3,4-tetrahydroisoquinoline (2) in an approx. 3:2 ratio. The prevalent formation of 1 over 2 reflects stereoselective cyclisation of a transient Schiff base in accord with the Felkin-Anh model. Fe3+ ions, present at relatively high levels in parkinsonian brains, markedly accelerated formation of 1 and 2. whereas Cu2+ decreased the reaction rate, due apparently to different sites of chelate formation between L-DOPA and the metal ions. Both metal ions markedly decreased the stereoselectivity of the reaction. Product 1 exhibited chelating properties toward metal ions comparable or stronger than those of L-DOPA. These results throw new light on the effects of transition metal ions on the Pictet-Spengler reaction

and suggest a possible role of tetrahydroisoquinoline products from L-DOPA and carbohydrate metabolites in the severe side effects of the drug. (C) 2001 Elsevier Science Ltd. All rights reserved.

Malecki EA. 2001. Manganese toxicity is associated with mitochondrial dysfunction and DNA fragmentation in rat primary striatal neurons. Brain Res Bull 55 (2):225-228.
Abstract: Manganese (Mn) in excess is toxic to neurons of the globus pallidus, leading to a Parkinsonian-like syndrome. We used rat primary neuron cultures to examine the cellular events following manganese exposure. Following exposure to Mn2+ for 48 h, striatal neurons showed dose-dependent losses of mitochondrial membrane potential and complex II activity. The Mn exposure effect on mitochondrial membrane potential was significant at every concentration measured (5, 50, and 500 muM), and the manganese exposure effect on complex II activity was significant at 50 and 500 muM. Exposure of striatal neurons to both Mn2+ and the complex II inhibitor 3-nitropropionic acid resulted in additive toxicity. Striatal neurons exposed to 5 muM Mn2+ for 48 h exhibited DNA fragmentation and decreases in the immunohistochemically detectable microtubule-associated protein MAP-2. These results indicate that manganese may trigger apoptotic-like neuronal death secondary to mitochondrial dysfunction. Rescue of neurons by apoptosis inhibitors may be helpful in treating manganese toxicity and similar neurodegenerative processes. (C) 2001 Elsevier Science Inc.

Louis ED. 2001. Etiology of essential tremor: Should we be searching for environmental causes? Mov Disord 16(5):822-829.

Loeffler DA, Sima AAF, Lewitt PA. 2001. Ceruloplasmin immunoreactivity in neurodegenerative disorders. Free Radic Res 35(2):111-118.
Abstract: Ceruloplasmin (CP) is a 132 kd cuproprotein which, together with transferrin, provides the majority of anti-oxidant capacity in serum. Increased iron deposition and lipid peroxidation in the basal ganglia of subjects with hereditary CP deficiency suggest that CP may serve as an anti-oxidant in the brain as well. The present study compared CP immunoreactivity in brain specimens from normal controls and subjects with neurodegenerative disorders (Alzheimer's disease [AD], Parkinson's disease [PD], progressive supranuclear palsy [PSP], and Huntington's disease [HD]) (n = 5 per group). The relative intensity of neuronal CP staining and the numbers of CP-stained neurons per 25x microscope field were determined in hippocampus (CAI, subiculum, and parahippocampal gyrus), parietal cortex, frontal cortex, substantia nigra, and caudate. CP was detected in both neurons and astrocytes in all specimens, and in senile plaques and occasional neurofibrillary tangles in AD brain. Neuronal CP staining intensity tended to increase in most AD brain regions, but was statistically significant vs controls only in the CAI region of hippocampus (p = .016). Neuronal CP staining in brain specimens from other neurodegenerative disorders showed a slight but nonsignificant increase vs controls. The numbers of CP-stained neurons per field did not differ between the various neurodegenerative disorders and controls. These results suggest that a modest increase in neuronal CP content is present in the AD brain, and lesser elevations in neuronal CP occur in the other neurodegenerative disorders in this study. Though CP functions as both an acute phase protein and an anti-oxidant in peripheral tissues, whether it does so in the brain remains to be determined.

Linder MC. 2001. Copper and genomic stability in mammals. Mutation Research-Fundamental and Molecular Mechanisms of Mutagenesis 475(1-2. Sp. Iss. Si): 141-152.
Abstract: As the free ion and in the form of some complexes, there is no doubt that copper can promote damage to cellular molecules and structures through radical formation. At the same time, and perhaps as a consequence? mammals have evolved means of minimizing levels of free copper ions and destructive copper complexes that enter the organism and its cells. These means include tight binding of copper ions to protein

carriers and transporters; direct exchange of copper between protein carriers, transporters, and cuproenzymes; and mobilization of secretory mechanisms and excretory pathways, as needed. As a consequence, normally, and except under certain genetic conditions, copper is likely to be benign to most mammals and not responsible for genomic instability, including fragmentation of and/or alterations to DNA, induction of mutations or apoptosis, or other toxic events. Indeed, cuproenzymes are important members of the antioxidant system of the organism. (C) 2001 Published by Elsevier Science B.V.

Lin E, Graziano JH, Freyer GA. 2001. Regulation of the 75-kDa subunit of mitochondrial complex I by iron. J Biol Chem 276(29):27685-27692.
Abstract: Iron homeostasis is tightly regulated, as cells work to conserve this essential but potentially toxic metal, The translation of many iron proteins is controlled by the binding of two cytoplasmic proteins, iron regulatory protein 1 and 2 (IRP1 and IRP2) to stem loop structures, known as iron-responsive elements (IREs), found in the untranslated regions of their mRNAs. In short, when iron is depleted, IRP1 or IRP2 bind IREs; this decreases the synthesis of proteins involved in iron storage and mitochondrial metabolism (e.g. ferritin and mitochondrial aconitase) and increases the synthesis of those involved in iron uptake (e.g. transferrin receptor). It is likely that more iron containing proteins have IREs and that other IRPs may exist. One obvious place to search is in Complex I of the mitochondrial respiratory chain, which contains at least 6 iron-sulfur (Fe-S) subunits, Interestingly, in idiopathic Parkinson's disease, iron homeostasis is altered, and Complex I activity is diminished. These findings led us to investigate whether iron status affects the Fe-S subunits of Complex I. We found that the protein levels of the 75-kDa subunit of Complex I were modulated by levels of iron in the cell, whereas mRNA levels were minimally changed. Isolation of a clone of the 75-kDa Fe-S subunit with a more complete 5'-untranslated region sequence revealed a novel IRE-like stem loop sequence. RNA protein gel shift assays demonstrated that a specific cytoplasmic protein bound the novel IRE and that the binding of the protein was affected by iron status. Western blot analysis and supershift assays showed that this cytosolic protein is neither IRP1 nor IRP2. In addition, ferritin IRE was able to compete for binding with this putative IRP, These results suggest that the 75-kDa Fe-S subunit of mitochondrial Complex I may be regulated by a novel IRE-IRP system.

Lin AMY. 2001. Coexistence of zinc and iron augmented oxidative injuries in the nigrostriatal dopaminergic system of SD rats. Free Radic Biol Med 30(3): 225-231.
Abstract: Clinical studies have demonstrated an excess of transition metals, including zinc and iron, in the substantia nigra (SN) of Parkinson's patients. In the present study, the neurotoxic effect of zinc was investigated using iron as a positive control. Addition of zinc or iron to brain homogenates increased lipid peroxidation. Zinc was less potent than iron in inducing lipid peroxidation. Coincubation with desferrioxamine prevented zinc- and iron-induced lipid peroxidation . Furthermore, glutathione (GSH), S-nitroso-N-acetylpenicillamine, or melatonin inhibited zinc-induced lipid peroxidation. The oxidative effect of zinc was further investigated in anesthetized rats. Seven days after intranigral infusion of zinc, lipid peroxidation was elevated in the infused SN, and dopamine content and tyrosine hydroxylasepositive axons were decreased in the ipsilateral striatum. Zinc was less potent than iron in inducing neurodegeneration in vivo. L-Buthionine-[S,R]-sulfoximine pretreatment (i.c.v.), which depletes cellular GSH levels, enhanced zinc induced oxidative injuries in the nigrostriatal doparninergic system. Moreover, simultaneous infusion of zinc and iron appeared to augment oxidative injuries in rat brain. Taken together, our results demonstrate that intranigral infusion of zinc caused degeneration of the nigrostriatal dopaminergic system in rat brain. Furthermore, coexistence of zinc and iron augmented oxidative injuries in rat brain. These findings indicate that both zinc and iron contribute to the etiology of Parkinsonism. (C) 2001

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Li SW, Lin TS, Minteer S, Burke WJ. 2001. 3,4-Dihydroxyphenylacetaldehyde and hydrogen peroxide generate a hydroxyl radical: possible role in Parkinson's disease pathogenesis. Molecular Brain Research 93(1):1-7.
Abstract: 3,4-Dihydroxyphenylacetaldehyde (DOPAL) and 3,4-dihydroxyphenylglycolaldehyde (DOPEGAL), the monoamine oxidase (MAO) metabolites of dopamine (DA) and norepinephrine (NE), respectively, are toxic to catecholamine (CA) neurons in vitro and in vivo. DOPEGAL generates a free radical and activates mitochondrial permeability transition, a mechanism implicated in neuron death. To determine if DOPAL and other DA metabolites generate the hydroxyl radical in the presence of H2O2, we used HPLC-EC to detect salicylate hydroxylation products. To determine the relative reducing capacity of DOPAL and DOPEGAL we used cyclic voltammetry to measure their reduction potentials. Results indicate that DOPAL, but not DOPEGAL, DA or other DA metabolites. generates hydroxyl radicals. Atomic absorption spectroscopy and heavy metal screening indicate that this result is not due to contamination of DOPAL with iron or other heavy metals. DOPAL reduction potential (161 mV) is lower than that of DOPEGAL (235 mV). DOPAL is present in human substantia nigra. The implications of these findings to CA neuronal death in degenerative brain diseases are discussed. (C) 2001 Elsevier Science B.V. All rights reserved.

Levites Y, Weinreb O, Maor G, Youdim MBH, Mandel S. 2001. Green tea polyphenol (-)-epigallocatechin-3-gallate prevents N-methyl-4-phenyl-1,2,3,6-tetrahydropyridine-induced dopaminergic neurodegeneration. J Neurochem 78(5):1073-1082.
Abstract: In the present study we demonstrate neuroprotective property of green tea extract and (-)-epigallocatechin-3-gallate in N-methyl-4-phenyl-1,2,3,6-tetrahydropyridine mice model of Parkinson's disease. N-Methyl-4-phenyl-1,2,3,6-tetrahydropyridine neurotoxin caused dopamine neuron loss in substantia nigra concomitant with a depletion in striatal dopamine and tyrosine hydroxylase protein levels. Pretreatment of mice with either green tea extract (0.5 and 1 mg/kg) or (-)-epigallocatechin-3-gallate (2 and 10 mg/kg) prevented these effects. In addition, the neurotoxin caused an elevation in striatal antioxidant enzymes superoxide dismutase (240%) and catalase (165%) activities, both effects being prevented by (-) -epigallocatechin-3-gallate. (-)-Epigallocatechin-3-gallate itself also increased the activities of both enzymes in the brain. The neuroprotective effects are not likely to be caused by inhibition of N-methyl-4-phenyl-1,2,3,6-tetrahydropyridine conversion to its active metabolite 1-methyl-4-phenylpyridinium by monoamine oxidase-B, as both green tea and (-)-epigallocatechin-3-gallate are very poor inhibitors of this enzyme in vitro (770 mug/mL and 660 mum, respectively). Brain penetrating property of polyphenols, as well as their antioxidant and iron-chelating properties may make such compounds an important class of drugs to be developed for treatment of neurodegenerative diseases where oxidative stress has been implicated.

Lee PL, Halloran C, Beutler E. 2001. Polymorphisms in the transferrin 5 ' flanking region associated with differences in total iron binding capacity: Possible implications in iron homeostasis. Blood Cells Mol Dis 27(2):539-548.
Abstract: We have identified five single nucleotide polymorphisms (SNPs) upstream (5 ') of the transferrin coding region. One polymorphism is in the 5 ' UTR ar nt +49, and four are in the promoter region at nt -34, -551, -617, and -739, numbering from the start of transcription. The -34 and -617 SNPs are tightly but not completely linked. The -34 polymorphism lies between a conserved Spl site and the TATA box. The -617 polymorphism is within the DRII enhancer region. Five haplotypes have been defined from these SNPs by the identification of at least one homozygous individual, and two other haplotypes were deduced from heterozygous individuals. The total iron-binding capacity associated with each transferrin haplotype was haplotype 2 > 1 > 4 > 3. Transferrin promoter haplotype 2 had a significantly higher mean TIBC and haplotype 3 had a significantly lower

mean TIBC than the more common haplotype 1. Persons with haplotype 4, which includes the -34T and -617A minor alleles, have a lower mean TIBC but the difference was not statistically significant. In normal individuals, the differences in the haplotypes were not found to be associated with differences in transferrin saturation and ferritin levels. There was no difference in the extent of increase in the mean TIBC levels in individuals with iron deficiency anemia in regard to their haplotype. Furthermore, there was no difference in the relative frequencies of the transferrin haplotypes in the iron-deficient population. In hemochromatosis patients who were homozygous for the C282Y HFE mutation, no particular haplotype was associated with a significant difference in transferrin saturation or ferritin levels. In White patients with Parkinson's disease, a disorder in which there is abnormal iron deposition in the brain, the presence of transferrin haplotype 3 was in slight excess over the normal White population. (C) 2001 Academic Press.

Lee D, Lee EK, Lee JH, Chang CS, Paik SR. 2001. Self-oligomerization and protein aggregation of alpha-synuclein in the presence of Coomassie Brilliant Blue. Eur J Biochem 268(2):295-301.
Abstract: alpha -Synuclein has been implicated in various neurodegenerative disorders, including Parkinson's and Alzheimer's diseases, by its participation in abnormal protein depositions. As the protein has been suggested to play a significant role in the formation of the deposits which might be responsible for neurodegeneration, there is a strong demand to screen for alpha -synuclein-interactive small molecules. In this report, Coomassie Brilliant Blue (CBB) interaction of alpha -synuclein has been investigated with respect to induction of protein self-oligomerization in the presence of the chemical coupling reagent N-(ethoxycarbonyl)-2-ethoxy-1,2-dihydroquinoline. Both CBB-G and CBB-R, which differ by only two methyl groups, induced the self-oligomerization of alpha -synuclein in a biphasic manner with optimal dye concentrations of 250 mum and 150 mum, respectively. The protein aggregates of alpha -synuclein induced by the dyes in the absence of the coupling reagent were analysed by electron microscopy. Whereas CBB-G induced formation of protein aggregates with a worm-like structure, CBB-R induced clear fibrilization of alpha -synuclein on a background of granular structures. CBB-R interacted with alpha -synuclein approximately twice as effectively as CBB-G (dissociation constants 0.63 mum and 1.37 mum, respectively). These dye interactions were independent from the acidic C-terminus of alpha -synuclein, which was reminiscent of the A beta 25-35 interaction of alpha -synuclein. However, the metal-catalysed oxidative self-oligomerization of alpha -synuclein in the presence of Cu2+/H2O2, which was augmented synergistically by A beta 25-35, was not affected by the dyes. This indicates that the dye binding site is also distinctive from the A beta 25-35 interaction site on alpha -synuclein. These biochemically specific interactions between alpha -synuclein and the dyes indicate that alpha -synuclein-interactive small molecules could provide a tool with which to approach development of diagnostic, preventive, or therapeutic strategies for various alpha -synuclein-related neurodegenerative disorders.

Lai GH, Chen CF, Su Y, Ho LT, Lin AMY. 2001. Lack of Protective Effect by Intermittent Hypoxia on Mptp-Induced Neurotoxicity in MiceVolume 939. p 33-44. Neuroprotective Agents: Annals of the New York Academy of Sciences.
Abstract: In contrast to acute ischemia and subsequent reperfusion that produce excess free radicals, intermittent hypoxia (III) is reported to play an important role in upregulation of antioxidative defensive mechanisms. In the study we report here, the neuroprotective effect of III was evaluated using intraperitoneal injection of 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) in ICR mice. Adult male ICR mice were subjected to 380 mmHg in an altitude chamber for 15 hours/day for 14 or 28 days. MPTP decreased striatal dopamine content in normoxic mice. However, IH did not significantly alter the MPTP-induced depletion of striatal dopamine content. Furthermore, MPTP had no effect on GSH

content but reduced GSH/GSSG ratio irt mouse striatum. III altered neither GSH content nor MPTP-induced reduction in GSH/GSSG ratio. Although MPTP had no effect on striatal SOD activity in normoxic mouse, III increased SOD activity in the saline and MPTP groups. Neither MPTP nor III affected GPx in mouse striatum. Furthermore, in our ex vivo study, both the autooxidation and iron induced lipid peroxidation of cortical homogenates were lower in the IH-treated group than those of the normoxic group, indicating a reduced oxidative status after III treatment. In conclusion, exposure to IH has been suggested to be beneficial in preventing iron-induced oxidative injuries in biological organisms, and our data support this notion in that III not only decreased iron-induced lipid peroxidation but also increased antioxidative defense enzyme activity in mouse brain. Furthermore, the lack of neuroprotective effect by Hi of MPTP-induced depletion of striatal dopamine content suggests that oxidative stress may not be the only mechanism for the MPTP-induced neurotoxicity.

Kunikowska G, Jenner P. 2001. 6-hydroxydopamine-lesioning of the nigrostriatal pathway in rats alters basal ganglia mRNA for copper, zinc- and manganese-superoxide dismutase, but not glutathione peroxidase. Brain Res 922(1):51-64.
Abstract: The effects of nigrostriatal pathway destruction on the mRNA levels of copper, zinc-dependent superoxide dismutase (Cu.Zn-SOD), manganese-dependent superoxide dismutase (Mn-SOD). and glutathione peroxidase in basal ganglia of adult rat were investigated using in situ hybridization histochemistry and oligodeoxynucleotide (single-stranded complementary DNA) probes. The 6-hydroxydopamine (6-OHDA)-induced destruction of the nigrostriatal pathway resulted in contralateral rotation to apomorphine and a marked loss of specific [H-3]mazindol binding in the striatum (93%; P <0.05) and of tyrosine hydroxylase mRNA in substantia nigra pars compacta (SC) (93%, P <0.05) compared with control rats. Levels of Cu,Zn-SOD rnRNA were decreased in the striatum, globus pallidus, and SC on the lesioned side of 6-OHDA-lesioned rats compared with sham-lesioned rats (P <0.05). Levels of Mn-SOD mRNA were increased in the nucleus accumbens (P <0.05), but decreased in the SC (P <0.05) on the lesioned side of 6-OHDA-treated rats compared with sham-lesioned rats. Lesioning with 6-OHDA had no effect on glutathione peroxidase mRNA levels in any region of basal ganglia examined. The significant changes in Cu,Zn-SOD and Mn-SOD rnRNA indicate that SOD is primarily expressed by dopaminergic neurons of the nigrostriatal pathway, and that the Mn-SOD gene appears to be inducible in rat basal ganglia in response to both physical and chemical damage 5 weeks after 6-OHDA-lesioning. These findings may clarify the status of antioxidant enzymes, particularly Mn-SOD. in patients with Parkinson's disease and their relevance to disease pathogenesis. (C) 2001 Published by Elsevier Science B.V.

Krieger J, Schroeder C. 2001. Iron, brain and restless legs syndrome. Sleep Medicine Reviews 5(4):277-286.
Abstract: Iron is the most important transitional metal in the body, as it is implicated in many metabolic processes, mostly related to its capacity as an electron donor/acceptor. Iron deficiency has been long been known to cause anaemia, iron excess to cause haemochromatosis. As excess free iron can cause oxidative damage, it is important that the levels of iron in the body are tightly regulated which appears to be done only by digestive absorption, as there is no known regulating mechanism for elimination of iron. The amount of free iron is also kept to a minimum thanks to binding to transferrin for transport, and to ferritin for storage. Recent research has put emphasis on the possible role of excess iron in the brain in several degenerative diseases. Iron deficiency in the central nervous system is known to cause motor impairment and cognitive deficits; more recently, it has been suggested that it may play a role in the pathophysiology of the restless leg syndrome. (C) 2001 Harcourt Publishers Ltd.

Kimura S, Kurasaki M, Saito T, Ito K, Hosokawa T, Okabe M, Shiraishi K, Niioka

T. 2001 . Effects of synthetic dopamine-melanins on oxygen radical formation induced by metal ions with dopamine. Neuroscience Research Communications 29(1):31-40.
Abstract: To understand the possible physiological roles of the neuromelanins, we examined effects of synthetic dopamine-melanins on oxygen radical formation by metal ions with dopamine. Oxygen radical formation was detected by DNA cleavage analysis. Addition of metal-free synthetic dopamine-melanins inhibited DNA cleavage induced by Fe or Cu ions with dopamine. On the other hand, in the presence of dopamine, dopamine-melanins loaded with Cu and Fe induced DNA cleavage without addition of free metal ions. It was clarified that dopamine-melanins loaded with Cu and Fe generated hydroxyl radicals in the presence of dopamine, on the basis of the findings that catalase and mannitol inhibited the DNA cleavage. Our results suggest that metal-free neuromelanins inhibit hydroxyl radical formation induced by metal ions with dopamine by sequestering metal ions.

Kim YS, Lee D, Lee EK, Sung JY, Chung KC, Kim J, Paik SR. 2001. Multiple ligand interaction of alpha-synuclein produced various forms of protein aggregates in the presence of A beta 25-35, copper, and eosin. Brain Res 908(1):93-98.
Abstract: Various protein aggregates of alpha -synuclein developed by way of the common protein self-oligomerization in the presence of A beta 25-35, copper, and eosin were examined. All the aggregates exhibited congo red birefringence although the actual amounts of the aggregates were varied as determined by thioflavin T binding fluorescence. When their morphologies were analyzed in relation to in vitro cytotoxicity, the smallest granular aggregates obtained with copper exhibited the highest cytotoxicity, while the fibrous structures by eosin did not affect the cell. (C) 2001 Elsevier Science B.V. All rights reserved.

Kim HJ, Soh Y, Jang JH, Lee JS, Oh YJ, Surh YJ. 2001. Differential cell death induced by salsolinol with and without copper: Possible role of reactive oxygen species. Mol Pharmacol 60(3):440-449.
Abstract: Salsolinol (SAL), a novel dopaminergic catechol tetrahydroisoquinoline neurotoxin, has been speculated to contribute to the etiology of Parkinson's disease and neuropathology of chronic alcoholism. Our previous studies have demonstrated that SAL induces strand scission in oX174 supercoiled DNA and oxidative base modification in calf thymus DNA in the presence of cupric ion. We now report that treatment of rat pheochromocytoma (PC12) cells with SAL causes reduced viability, which was exacerbated by Cu2+. The copper chelator bathocuproinedisulfonic acid ameliorated cytotoxicity induced by SAL and Cu2+. N-Acetyl-L-cysteine and reduced glutathione protected against SAL- plus Cu2+-mediated PC12 cell death. Cells exposed to SAL underwent apoptosis, as revealed by characteristic morphological and biochemical changes. SAL treatment resulted in increased levels of Bax with a concomitant decrease in expression of Bcl-X-L. Furthermore, SAL rapidly activated c-Jun N-terminal kinase, whereas the activity of extracellular signal-regulated protein kinase remained unchanged. Transfection with Bcl-X-L or Bcl-2 led to protection against SAL-mediated PC12 cell death. Although SAL alone could cause apoptotic death in PC12 cells, cells treated with SAL together with Cu2+ became necrotic. Cells exposed to both SAL and Cu2+ exhibited higher levels of intracellular reactive oxygen species, malondialdehyde, and 8-oxo-7,8-dihydro-2'-deoxyguanosine than did those treated with SAL alone. These results suggest that copper accelerates redox cycling of SAL, leading to massive production of reactive oxygen species, which can divert the SAL-induced cell death to necrosis.

Khan FH, Saha M, Chakrabarti S. 2001. Dopamine induced protein damage in mitochondrial-synaptosomal fraction of rat brain. Brain Res 895(1-2): 245-249.
Abstract: Dopamine during in vitro oxidation induced covalent cross-linking of membrane proteins in rat brain crude mitochondrial-synaptosomal fraction. The process is not inhibited by hydroxyl radical scavengers, lipid

soluble anti-oxidants, metal-chelator or catalase, but reduced glutathione produced a dramatic inhibition of cross-linking. The protein cross-linking mediated by dopamine is not associated with any detectable membrane lipid peroxidation but significant formation of protein bound quinone takes place during incubation. Our results indicate that reactive quinones rather than oxygen free radicals are involved in dopamine induced protein cross-linking in rat brain membrane fraction. (C) 2001 Elsevier Science B.V. All rights reserved.

Kahle PJ, Neumann M, Ozmen L, Muller V, Odoy S, Okamoto N, Jacobsen H, Iwatsubo T, Trojanowski JQ, Takahashi H, Wakabayashi K, Bogdanovic N, Riederer P, Kretzschmar HA, Haass C. 2001. Selective insolubility of alpha-Synuclein in human Lewy body diseases is recapitulated in a transgenic mouse model. Am J Pathol 159(6):2215-2225.
Abstract: alpha -Synuclein (alpha -SYN) is deposited in intraneuronal cytoplasmic inclusions (Lewy bodies, LBs) characteristic for Parkinson's disease (PD) and LB dementias. alpha -SYN forms LB-like fibrils in vitro, in contrast to its homologue beta -SYN. Here we have investigated the solubility of SYNs in human LB diseases and in transgenic mice expressing human wild-type and PD-associated mutant [A30P]alpha -SYN driven by the brain neuron-specific promoter, Thy1. Distinct alpha -SYN species were detected in the detergent-insoluble fractions from brains of patients with PD, dementia with LBs, and neurodegeneration with brain iron accumulation type 1 (formerly known as Hallervorden-Spatz disease). Using the same extraction method, detergent-insolubility of human alpha -SYN was observed in brains of transgenic mice. In contrast, neither endogenous mouse alpha -SYN nor beta -SYN were detected in detergent-insoluble fractions from transgenic mouse brains. The nonamyloidogenic beta -SYN was incapable of forming insoluble fibrils because amino acids 73 to 83 in the central region of alpha -SYN are absent in beta -SYN. In conclusion, the specific accumulation of detergent-insoluble alpha -SYN in transgenic mice recapitulates a pivotal feature of human LB diseases.

Jung YJ, Surh YJ. 2001. Oxidative DNA damage and cytotoxicity induced by copper-stimulated redox cycling of salsolinol, a neurotoxic tetrahydroisoquinoline alkaloid. Free Radic Biol Med 30(12):1407-1417.
Abstract: A series of neurotoxic tetrahydroisoquinoline alkaloids has been detected in certain regions of mammalian brains. One such dopaminergic tetrahydroisoquinoline neurotoxin is salsolinol (SAL), which is suspected of being associated with the etiology of Parkinson's disease and neuropathology of chronic alcoholism. In the present study, we found that SAL in combination with Cu(II) induced strand scission in pBR322 and phi X174 supercoiled DNA, which was inhibited by the copper chelator, reactive oxygen species (ROS) scavengers, reduced glutathione, and catalase. SAL in the presence of Cu(II) caused hydroxylation of salicylic acid to produce 2,3- and 2,5-dihydroxybenzoic acids. Reaction of calf thymus DNA with SAL plus Cu(II) resulted in substantial oxidative DNA damage as determined by 8-hydroxydeoxyguanosine (8-OH-dG) formation. Blockade of the dihydroxy functional group of SAL abolished its capability to yield 8-OH-dG in the presence of Cu(LI). The dehydro analog of SAL, 1-methyl-6,7-dihydroxy-3,4dihydroisoquinoline, produced significantly high levels of 8-OH-dG when incubated with calf thymus DNA, even in the absence of Cu (II), which appears to be attributable to the tautomer formation by this compound. In another experiment, SAL exerted cytotoxicity when treated to rat pheochromocytoma (PC12) cells. Based on these findings, it seems likely that SAL undergoes redox cycling in the presence of Cu(II) with concomitant production of ROS, particularly hydroxyl radical, which could contribute to DNA damaging and cytotoxic properties of this neurotoxin. (C) 2001 Elsevier Science Inc.

Johnson S. 2001. Micronutrient accumulation and depletion in schizophrenia, epilepsy, autism and Parkinson's disease? Med Hypotheses 56(5):641-645.
Abstract: Zinc has several crucial functions in brain development and maintenance: it binds to p53, preventing it from binding to supercoiled DNA and ensuring that p53 cause the expression of several paramount genes,

such as the one that encodes for the type I receptors to pituitary adenine cylase-activator peptide (PACAP), which directs embryonic development of the brain cortex, adrenal glands, etc.; it is required for the production of CuZnSOD and Zn-thionein, which are essential to prevent oxidative damage; it is required for many proteins, some of them with Zn fingers, many of them essential enzymes for growth and homeostasis. For example, the synthesis of serotonin involves Zn enzymes and since serotonin is necessary for melatonin synthesis, a Zn deficiency may result in low levels of both hormones. Unfortunately, Zn levels tend to be low when there is excess Cu and Cd. Moreover, high estrogen levels tend to cause increased absorption of Cu and Cd, and smoking and eating food contaminated with Cd result in high levels of the latter. Furthermore, ethanol ingestion increases the elimination of Zn and Mg (which acts as a cofactor for CuZnSOD). Increased Cu levels may also be found in people with Wilson's disease, which is a rather rare disease. However, the heterozygote form (only one faulty copy of the chromosome) is not so rare. Therefore, the developing fetus of a pregnant women who is low in Zn and high in Cu may experience major difficulties in the early development of the brain, which may later manifest themselves as schizophrenia, autism or epilepsy. Similarly, a person who gradually accumulates Cu, will tend to experience a gradual depletion of Zn, with a corresponding increase in oxidative damage, eventually leading to Parkinson's disease. Also discussed are the crucial roles of histidine, histamine, vitamin D, essential fatty acids, vitamin E, peroxynitrate, etc. in the possible oxidative damage involved in these mental diseases. (C) 2001 Harcourt Publishers Ltd.

Johnson S. 2001. Is Parkinson's disease the heterozygote form of Wilson's disease: PD=1/2 WD? Med Hypotheses 56(2):171-173.
Abstract: Wilson's disease (WD) patients often present with Parkinson's disease (PD). Furthermore, most patients with PD have reduced ceruloplasmin, a characteristic of Wilson's disease. WD is an autosomal recessive disease (requires two faulty copies of a gene to produce a homozygote individual) that afflicts 1 in 1000 people. However, the number of people with one faulty copy (heterozygotes) is much larger, probably about 2% of the population. I hypothesize that the large number of heterozygotes for WD are at greatly increased risk for idiopathic PD, because these people accumulate free copper in the basal ganglia at a slower rate than homozygotes, which accounts for the fact that PD usually develops after 40 years of age. In WD, a ceruloplasmin deficiency results in accumulation of free Cu in the liver, brain, kidneys, etc. The excess Cu results in impaired Zn absorption, which would account for the low levels of Zn in the brains of PD patients. Moreover, the high levels of Fe found in the substantia nigra of PD patients may perhaps be explained by free Cu binding to iron binding protein-1 (IBP-1), causing it to malfunction and preventing it from detaching itself from the transferrin receptor (TfR) inhibition gene, resulting in expression of TfR even when the cell has plenty of Fe. The gradual accumulation of Fe and Cu would explain the damage inflicted on the substantia nigra by free radicals catalyzed by these two metals and which is exacerbated by the low levels of CuZnSOD, due to the Zn deficiency mentioned above. Moreover, if this hypothesis is correct, then PD could be used to help discover the gene (or genes) responsible for WD and vice versa. Furthermore, idiopathic PD could be prevented by identifying the heterozygote individuals and providing them with Zn supplementation, Cu chelation therapy and phlebotomy to eliminate Fe. (C) 2001 Harcourt Publishers Ltd.

Jensen JH, Chandra R, Yu H. 2001. Quantitative model for the interecho time dependence of the CPMG relaxation rate in iron-rich gray matter. Magn Reson Med 46(1):159-165.
Abstract: A quantitative model is proposed for computing the dependence on the interecho time of the NMR relaxation rate in iron-rich gray matter obtained with a Carr-Purcell-Meiboom-Gill sequence. The model consists of representing oligodendrocytes as identical magnetic spheres arranged in a spatially random pattern, and in approximating water diffusion as isotropic

and unrestricted. Predictions of the model are calculated numerically using a Monte Carlo technique and, for the weak field limit, using an analytic formula. The model is shown to provide a good fit to experimental measurements of in vitro samples of monkey brain at field levels of 1.0 T and 1.5 T, These field levels are not sufficient to fully determine the model parameters, but it is argued that this may be possible at 3.0 T, The model is potentially of value for multiple-spin-echo MRI studies of iron-related neurodegenerative disorders, such as Parkinson's disease. In particular, the model can be applied to correlate MRI data with the cellular distribution of iron in gray matter. Magn Reson Med 46:159-165, 2001. (C) 2001 Wiley-Liss, Inc.

Jameson GNL, Linert W. 2001. The oxidation of 6-hydroxydopamine in aqueous solution. Part 2. Speciation and product distribution with iron(III) as oxidant. Journal of the Chemical Society-Perkin Transactions 2 (4): 563-568.
Abstract: 6-Hydroxydopamine [5-(2-aminoethyl)benzene-1,2,4-triol, protonated form H3LH+] reacts anaerobically with aqueous iron(III) to produce four types of quinone, namely the p-(pQ), o-(oQ), triketo- (tQ) and the deprotonated quinone (Q(-)). The relative concentrations depend strongly on the pH. At low pH pQ, oQ and tQ predominate and are in mete-stable equilibrium. However, the pH dependency of the concentrations of pQ and tQ at low pH suggests an equilibrium between their respective semiquinones. Above a pH of about 2, tQ disappears and above about 2.5 pQ and oQ equilibrate via Q(-), which, above a pH of about 6, is the only species present. The equilibrium constants and molar absorbances have been measured for all these species and are discussed. The speciation over the pH range 0.5-5 has been determined as a necessary prelude to a full kinetic study of the system.

Imam SZ, El-Yazal J, Newport GD, Itzhak Y, Cadet JL, Slikker W, Ali SF. 2001. Methamphetamine-Induced Dopaminergic Neurotoxicity: Role of Peroxynitrite and Neuroprotective Role of Antioxidants and Peroxynitrite Decomposition CatalystsVolume 939. p 366-380. Neuroprotective Agents: Annals of the New York Academy of Sciences.
Abstract: Oxidative stress, reactive oxygen (ROS), and nitrogen (RNS) species have been known to be involved in a multitude of neurodegenerative disorders such as Parkinson's disease (PD), Alzheimer's disease (AD), and amyotrophic lateral sclerosis (ALS). Both ROS and RNS have very short half-lives, thereby making their identification very difficult as a specific cause of neurodegeneration. Recently, we have developed a high performance liquid chromatography/electrochemical detection (HPLC/EC) method to identify 3-nitrotyrosine (3-NT), an in vitro and in vivo biomarker of peroxynitrite production, in cell cultures and brain to evaluate if an agent-driven neurotoxicity is produced by the generation of peroxynitrite. We show that a single or multiple injections of methamphetamine (METH) produced a significant increase in the formation of 3-NT in the striatum. This formation of 3-NT correlated with the striatal dopamine depletion caused by METH administration. We also show that PC12 cells treated with METH has significantly increased formation of 3-NT and dopamine depletion. Furthermore, we report that pretreatment with antioxidants such as selenium and melatonin can completely protect against the formation of 3-NT and depletion of striatal dopamine. We also report that pretreatment with peroxynitrite decomposition catalysts such as 5,10,15,20-tetrakis(N-methyl-4'-pyridyl)porphyrinato iron III (FeTMPyP) and 5, 10, 15, 20-tetrakis (2,4,6-trimethyl-3,5-suffonatophenyl) porphinato iron III (FETPPS) significantly protect against METH-induced 3-NT formation and striatal dopamine depletion. We used two different approaches, pharmacological manipulation and transgenic animal models, in order to further investigate the role of peroxynitrite. We show that a selective neuronal nitric oxide synthase (nNOS) inhibitor, 7-nitroindazole (7-NI), significantly protect against the formation of 3-NT as well as striatal dopamine depletion. Similar results were observed with nNOS knockout and copper zinc superoxide dismutase (CuZnSOD)-overexpressed transgenic mice models. Finally, using the protein data bank crystal

structure of tyrosine hydroxylase, we postulate the possible nitration of specific tyrosine moiety in the enzyme that can be responsible for dopaminergic neurotoxicity. Together, these data clearly support the hypothesis that the reactive nitrogen species, peroxynitrite, plays a major role in METH-induced dopaminergic neurotoxicity and that selective antioxidants and peroxynitrite decomposition catalysts can protect against METH-induced neurotoxicity. These antioxidants and decomposition catalysts may have therapeutic potential in the treatment of psychostimulant addictions.

Hudson NJ, Evans AT, Yeung CK, Hewitt PJ. 2001. Effect of process parameters upon the dopamine and lipid peroxidation activity of selected MIG welding fumes as a marker of potential neurotoxicity. Ann Occup Hyg 45(3): 187-192.
Abstract: There is growing concern over the neurotoxic effects of chronic occupational exposure to metal fume produced by welding. Elevated iron and manganese levels in the brain have been linked to an increase in lipid peroxidation, dopamine depletion and predisposition to the development of a Parkinson's type condition in advanced cases. Chemical and toxicological analysis of selected welding fumes, generated by model processes, were used in order to evaluate their potential to release solutes that promote oxidation of dopamine and peroxidation of brain lipids in cell free assays, This study compared the effect of shield gas, electrode type and voltage/ currect upon the dopamine and brain lipid peroxidation potential of selected welding fume, obtained from metal inert gas (MIG) welding systems, Overall, fume extracts were found to enhance dopamine oxidation and inhibit lipid peroxidation. Significant differences were also found in the oxidising potential of fume generated under differing process conditions; it may therefore be possible to determine the potential neurotoxicity of fumes using this system. (C) 2001 British Occupational Hygiene Society. Published by Elsevier Science Ltd. All rights reserved.

Hirata Y, Kiuchi K, Nagatsu T. 2001. Manganese mimics the action of 1-methyl-4-phenylpyridinium ion, a dopaminergic neurotoxin, in rat striatal tissue slices. Neurosci Lett 311(1):53-56.
Abstract: Manganese and 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) are known to induce neurological pathologies similar to that of parkinsonism. Previous studies performed in rat striatal slices have shown that MPTP and related compounds inhibit tyrosine hydroxylation, a rate-limiting step of dopamine biosynthesis. Here, we reported that manganese inhibited tyrosine hydroxylation in rat striatal slices. In addition, manganese caused increase in the levels of lactate indicating that aerobic glycolysis was inhibited in striatal slices. This inhibition was unique to manganese since other divalent cations, such as magnesium and zinc, did not increase lactate concentrations. These results suggest that the mechanisms by which manganese produces dysfunction of the nervous system are similar to those of MPTP. (C) 2001 Elsevier Science Ireland Ltd. All rights reserved.

Herrera F, Sainz RM, Mayo JC, Martin V, Antolin I, Rodriguez C. 2001. Glutamate induces oxidative stress not mediated by glutamate receptors or cystine transporters: protective effect of melatonin and other antioxidants. J Pineal Res 31(4):356-362.
Abstract: Glutamate is responsible for most of the excitatory synaptic activity and oxidative stress induction in the mammalian brain. This amino acid is increased in the substantia nigra in parkinsonism due to the lack of dopamine restraint to the subthalamic nucleus. Parkinson's disease also shows an increase of iron levels in the substantia nigra and a decrease of glutathione, the antioxidant responsible for the ascorbate radical recycling. Considered together, these facts could make the antioxidant ascorbate behave as a pro-oxidant in parkinsonism. Since both glutamate and ascorbate are present in the synaptosomes and neurons of substantia nigra, we tested 1) if glutamate is able to induce oxidative stress independently of its excitatory activity, and 2) if ascorbate may have synergistic effects with glutamate when these two molecules co-exist.

Brains were homogenized in order to disrupt membranes and render membrane receptors and intracellular signaling pathways non-functional. In these homogenates glutamate induced lipid peroxidation, indicating that this amino acid also may cause oxidative stress not mediated by its binding to glutamate receptors or cystine transporters. Ascorbate also induced lipid peroxidation thus behaving as a pro-oxidant. Both substances together produced an additive effect but they did not synergize. Given that melatonin is a potent physiological antioxidant with protective effects in models of neurotoxicity, we tested the role of this secretory product on the pro-oxidant effect of both compounds given separately or in combination. We also checked the protective ability of several other, antioxidants. Pharmacological doses of melatonin (millimolar), es estrogens, pinoline and trolox (micromolar) prevented the oxidant. effect of glutamate, ascorbate, and the combination of both substances. Potential therapeutic application of these results is discussed.

Heron P, Cousins K, Boyd C, Daya S. 2001. Paradoxical effects of copper and manganese on brain mitochondrial function. Life Sci 68 (14):1575-1583.
Abstract: Defects in the mitochondrial genome have been associated with Parkinson's and Alzheimer's disease, and apoptosis can be triggered by the presence of energetically compromised mitochondria. Thus, in this study we have examined whether the divalent cations CU2+ and Mn2+ could influence mitochondrial function ia vitro. Mitochondrial electron transport was dose and time dependently reduced by CU2+ to a greater extent with succinate as a substrate. Following a 60min preincubation period, Mn2+ dose dependently inhibited electron transport to a greater extent with lactate and malate. In contrast, paradoxical effects were seen following a 5min preincubation period with Mn2+. CU2+ dose-dependently reduced NADH-dependent lactate dehydrogenase (LDH) activity, with almost complete inhibition apparent at 10 muM. An initial induction of LDH by 10 muM Mn2+ was partially reversed by higher concentrations of the metal. CU2+ dose-dependently reduced flavin adenine dinucleotide (FAD) dependent monoamine oxidase A (MAO-A) activity in a time-independent manner, with an IC50 value approximate to 20 muM, whereas Mn2+ had no effect. In conclusion, it is proposed that CU2+ and Mn2+ have differential effects on nicotinamide adenine dinucleotide (NAD) and FAD-dependent mitochondrial enzymes at the level of the essential cofactors. CU2+ appears to exert an inhibitory effect on both NAD and FAD-dependent enzymes, but predominantly against the latter, including MAO-A and succinate dehydrogenase. The complex responses to Mn2+ may be due to dose-related effects on the interconversion of NAD and NADH and reversible enzymatic reactions employing this nucleotide cofactor. (C) 2001 Elsevier Science Inc. All rights reserved.

Hayes J, Tipton KF, Bianchi L, Della Corte L . 2001. Complexities in the neurotoxic actions of 6-hydroxydopamine in relation to the cytoprotective properties of taurine. Brain Res Bull 55(2):239-245.
Abstract: The neurotoxin 6-hydroxydopamine was shown to cause an imbalance between the direct and indirect pathways of the striato-nigral system as evidenced by a decreased release of gamma -aminobutyric acid and taurine in the substantia nigra but not in the globus pallidus following neostriatal stimulation with kainate (100 muM). The neurotoxicity of 6-hydroxydopamine is generally believed to result from reactive-oxygen radical formation, although it is also known to inhibit mitochondrial NADH dehydrogenase. The release of Fe(ll) from the unactivated form [3Fe (III)-4S] of cytoplasmic aconitase (EC50 < 8 <mu>M) was shown to be followed by the slower oxidation of thiol groups in the protein. Complete loss of -SH groups, and enzyme activity, was seen after incubation of glyceraldenyde-3-phosphate dehydrogenase with 200 muM 6-hydroxydopamine for 75 min at 37 degreesC (IC50 = 70.8 +/- 0.3 muM). Thus the cellular effects of 6-hydroxydopamine are complex, involving impairment of mitochondrial function, iron- release, sulphydryl-group oxidation, and enzyme inhibition in addition to direct generation of reactive oxygen radicals. Taurine, which is known to be neuroprotective in some other systems, only affords protection against some of these effects,

thereby explaining its reported ineffectiveness against 6-hydroxydopamine toxicity. (C) 2001 Elsevier Science Inc.

Halliwell B. 2001. Role of free radicals in the neurodegenerative diseases - Therapeutic implications for antioxidant treatment. Drugs & Aging 18(9): 685-716.
Abstract: Free radicals and other so-called 'reactive species' are constantly produced in the brain in vivo. Some arise by 'accidents of chemistry', an example of which may be the leakage of electrons from the mitochondrial electron transport chain to generate superoxide radical (O-2(.-)). Others are generated for useful purposes, such as the role of nitric oxide in neurotransmission and the production Of O-2(.-) activated microglia. Because of its high ATP demand, the brain consumes O-2 rapidly, and is thus susceptible to interference with mitochondrial function, which can in turn lead to increased O-2(.-) formation. The brain contains multiple antioxidant defences, of which the mitochondrial manganese-containing superoxide dismutase and reduced glutathione seem especially important. Iron is a powerful promoter of free radical damage, able to catalyse generation of highly reactive hydroxyl, alkoxyl and peroxyl radicals from hydrogen peroxide and lipid peroxides, respectively. Although most iron in the brain is stored in ferritin, 'catalytic' iron is readily mobilised from injured brain tissue. Increased levels of oxidative damage to DNA, lipids and proteins have been detected by a range of assays in post-mortem tissues from patients with Parkinson's disease, Alzheimer's disease and amyotrophic lateral sclerosis, and at least some of these changes may occur early in disease progression. The accumulation and precipitation of proteins that occur in these diseases may be aggravated by oxidative damage, and may in turn cause more oxidative damage by interfering with the function of the proteasome. Indeed, it has been shown that proteasomal inhibition increases levels of oxidative damage not only to proteins but also to other biomolecules. Hence, there are many attempts to develop antioxidants that can cross the blood-brain barrier and decrease oxidative damage. Natural antioxidants such as vitamin E (tocopherol), carotenoids and flavonoids do not readily enter the brain in the adult, and the lazaroid antioxidant tirilazad (U-74006F) appears to localise in the blood-brain barrier. Other antioxidants under development include modified spin traps and low molecular mass scavengers Of O-2(.-). One possible source of lead compounds is the use of traditional remedies claimed to improve brain function. Little is known about the impact of dietary antioxidants upon the development and progression of neurodegenerative diseases, especially Alzheimer's disease. Several agents already in therapeutic use might exert some of their effects by antioxidant action, including selegiline (deprenyl), apomorphine and nitecapone.

Guo ZH, Lee J, Lane M, Mattson MP. 2001. Iodoacetate protects hippocampal neurons against excitotoxic and oxidative injury: involvement of heat-shock proteins and Bcl-2. J Neurochem 79(2):361-370.
Abstract: Mild metabolic stress may increase resistance of neurons in the brain to subsequent, more severe insults, as demonstrated by the ability of ischemic pre-conditioning and dietary restriction to protect neurons in experimental models of stroke- and age-related neurodegenerative disorders. In the present study we employed iodoacetic acid (IAA), an inhibitor of glyceraldehyde-3-phosphate dehydrogenase, to test the hypothesis that inhibition of glycolysis can protect neurons. Pre-treatment of cultured hippocampal neurons with IAA can protect them against cell death induced by glutamate, iron and trophic factor withdrawal. Surprisingly, protection occurred with concentrations of IAA (2-200 nm) much lower than those required to inhibit glycolysis. Pre-treatment with IAA results in suppression of oxyradical production and stabilization of mitochondrial function in neurons after exposure to oxidative insults. Levels of the stress heat-shock proteins HSP70 and HSP90, and of the anti-apoptotic protein Bcl-2, were increased in neurons exposed to IAA. Our data demonstrate that IAA can stimulate cytoprotective mechanisms within neurons, and suggest the possible use of IAA and related compounds in

the prevention and/or treatment of neurodegenerative conditions.

Grunblatt E, Mandel S, Maor G, Youdim MBH. 2001. Gene expression analysis in N-methyl-4-phenyl-1,2,3,6-tetrahydropyridine mice model of Parkinson's disease using cDNA microarray: effect of R-apomorphine. J Neurochem 78 (1):1-12.
Abstract: To establish the possible roles of oxidative stress, inflammatory processes and other unknown mechanisms in neurodegeneration, we investigated brain gene alterations in N-methyl-4-phenyl-1,2,3.6-tetrahydropyridine (MPTP) mice model of Parkinson's disease using Atlas mouse cDNA expression array membrane. The expression of 51 different genes involved in oxidative stress, inflammation, glutamate and neurotrophic factors pathways as well as in still undefined processes, such as cell cycle regulators and signal transduction molecules, was differentially affected by the treatment. The present study indicates the involvement of an additional cascade of events that might act in parallel to oxidative stress and inflammation to converge eventually into a common pathway leading to neurodegeneration. The attenuation of these gene changes by R-apomorphine, an iron chelator-radical scavenger drug, supports our previous findings in vivo where R-apomorphine was neuroprotective.

Grunblatt E, Mandel S, Maor G, Youdim MBH. 2001. Effects of R- and S-apomorphine on MPTP-induced nigro-striatal dopamine neuronal loss. J Neurochem 77(1):146-156.
Abstract: In order to establish whether the antioxidant and iron-chelating activities of R-apomorphine (R-APO), a D-1-D-2 receptor agonist, may contribute to its neuroprotective property, its S-isomer, which is not a dopamine agonist, was studied. The neuroprotective property of R- and S-APO has been studied in the MPTP model of Parkinson's disease (PD). Both S-APO (0.5-1 mg/kg, subcutaneous) and R-APO (10 mg/kg) pretreatment of C57-BL mice, protected against MPTP (24 mg/kg, intraperitoneally) induced dopamine (DA) depletion and reduction in tyrosine hydroxylase (TH) activity. However, only R-APO prevented nigro-striatal neuronal cell degeneration, as indicated by the immunohistochemistry of TH positive neurones in substantia nigra and by western analysis of striatal TH content. R-APO prevented the reduction of striatal-GSH and the increase in the ratio of GSSG over total glutathione, caused by MPTP treatment. In vitro both R-APO and S-APO inhibited monoamine oxidase A and B activity at relatively high concentrations (100 and 300 mu mol/L, respectively). The elevated activity of TH induced by the two enantiomers may contribute to the maintenance of normal DA levels, suggesting that one of the targets of these molecules may involve upregulation of TH activity. It is suggested that the antioxidant and iron-chelating properties, possible monoamine oxidase inhibitory actions, together with activation of DA receptors, may participate in the mechanism of neuroprotection by APO enantiomers against MPTP.

Grau AJ, Willig V, Fogel W, Werle E. 2001. Assessment of plasma lactoferrin in Parkinson's disease. Mov Disord 16(1):131-134.
Abstract: Iron may play an important role in the pathogenesis of Parkinson's disease (PD). Recent studies have shown that the iron-transporting glycoprotein lactoferrin (LF) and its receptor are increased in the substantia nigra (SN) in PD. We investigated whether plasma levels of LF are altered in dopa-responsive PD. Plasma LF was not different between patients with PD (n = 23: 306 +/- 116 [mean +/- standard deviation] ng/ml) and age- and sex-matched healthy control subjects(n = 15: 359 +/- 126 ng/ml). However, LF was inversely correlated with PD severity (r = -0.68. P = 0.002), an association that remained significant after adjustment for treatment with levodopa. monoaminooxidase inhibitors, and dopa agonists (r = -0.53 P = 0.017). Plasma transferrin and ferritin levels were not different between groups and neither correlated with disease severity nor with LF levels. Together with the result of increased nigral lactoferrin, this finding is compatible with the hypothesis of an imbalance between LF levels in blood and SN in progressing PD. Larger and particularly

longitudinal studies and measurements of LF in cerebrospinal fluid are warranted to further examine the role of LF in PD.

Graham JM, Paley MNJ, Grunewald RA, Hoggard N, Griffiths PD. 2001. Brain iron deposition in Parkinson's disease imaged using the PRIME magnetic resonance sequence (vol 123, pg 2423, 2000). Brain 124:1258.

Gilgun-Sherki Y, Melamed E, Offen D. 2001. Oxidative stress induced-neurodegenerative diseases: the need for antioxidants that penetrate the blood brain barrier. Neuropharmacology 40(8):959-975.
Abstract: Oxidative stress (OS) has been implicated in the pathophysiology of many neurological, particularly neurodegenerative diseases. OS can cause cellular damage and subsequent cell death because the reactive oxygen species (ROS) oxidize vital cellular components such as lipids, proteins, and DNA. Moreover, the brain is exposed throughout life to excitatory amino acids (such as glutamate), whose metabolism produces ROS, thereby promoting excitotoxicity. Antioxidant defense mechanisms include removal of O-2, scavenging of reactive oxygen/nitrogen species or their precursors, inhibition of ROS formation, binding of metal ions needed for the catalysis of ROS generation and up-regulation of endogenous antioxidant defenses. However, since our endogenous antioxidant defenses are not always completely effective, and since exposure to damaging environmental factors is increasing, it seems reasonable to propose that exogenous antioxidants could be very effective in diminishing the cumulative effects of oxidative damage. Antioxidants of widely varying chemical structures have been investigated as potential therapeutic agents. However, the therapeutic use of most of these compounds is limited since they do not cross the blood brain barrier (BBB). Although a few of them have shown limited efficiency in animal models or in small clinical studies, none of the currently available antioxidants have proven efficacious in a large-scale controlled study. Therefore, any novel antioxidant molecules designed as potential neuroprotective treatment in acute or chronic neurological disorders should have the mandatory prerequisite that they can cross the BBB alter systemic administration. (C) 2001 Elsevier Science Ltd. All rights reserved.

Friedman A, Galazka-Friedman J. 2001. The Current State of Free Radicals in Parkinson's Disease - Nigral Iron as a Trigger of Oxidative StressVolume 86. p 137-142. Parkinson's Disease: Advances in Neurology.

Fredriksson A, Schroder N, Eriksson P, Izquierdo I, Archer T. 2001. Neonatal iron potentiates adult MPTP-induced neurodegenerative and functional deficits. Parkinsonism & Related Disorders 7(2):97-105.
Abstract: The interactive effects of neonatal iron and adult MPTP treatment groups of C57 B1/6 mice were studied through adminustration of iron (Fe2 +) 7.5 mg/kg b.w., p.o. or vehicle (saline) on days 10-12 post partum, followed at 3 months of age by administration of either MPTP (2 x 20 or 2 x 40 mg/kg, s.c.) or saline. Neonatal iron administration to mice-induced hypoactivity during the first 20-min period of testing and hyperactivity during the 3rd and final 20-min period for all three parameters of motor activity tested at 4 months of age. MPTP treatment caused a dose-related hypokinesia throughout the 3 x 20-min test periods; in the mice that received both neonatal iron and MPTP severe deficits of motor activity (akinesia) were obtained. Iron treatment impaired the ability of mice to habituate to the novel testing environment and later administration of MPTP potentiated the impairment markedly. Neurochemical analyses of striatal and frontal cortical dopamine (DA) and DA metabolites demonstrated that the depletions were potentiated under conditions of combined neonatal iron and adult MPTP. The analysis of total iron content (mug/g) in brain regions indicated notably elevated levels in the basal ganglia, but not in the frontal cortex, of mice administered Fe2+. Iron-overload combined with MPTP treatment induced functional and neurochemical deficits with interactive consequences beyond a mere additive effect that may have implications for the neurodegenerative process in parkinsonism. (C) 2001 Elsevier

Science Ltd. All rights reserved.

Fok TF, Chui KKM, Cheung R, Ng PC, Cheung KL, Hjelm M. 2001. Manganese intake and cholestatic jaundice in neonates receiving parenteral nutrition: a randomized controlled study. Acta Paediatr 90(9):1009-1015.
Abstract: Infants requiring parenteral nutrition (n = 244) were randomized to receive either 1 (group 1, n = 121) or 0.0182 mu mol/kg/d (group 2, n = 123) of manganese supplementation. The whole-blood manganese and serum direct bilirubin concentrations of the infants were monitored, as was the development of cholestasis (peak serum direct bilirubin concentration > 50 mu mol/L). Subgroup analysis was carried out on the data of 78 infants in group I and 82 in group 2 who had received manganese supplementation and more than three-quarters of their total daily fluid as parenteral nutrition for > 14 d. Of all the infants randomized, the high manganese group (group 1) showed a trend towards developing higher peak whole-blood manganese concentration [group I versus group 2: median (interquartile range): 606.0 (421.0; 1005.0) vs 566.0 (336.0; 858.0); p=0.061] and higher peak serum direct bilirubin concentration [37.0 (10.5; 122.5) vs 19.0 (8.0, 112.5); p = 0.153], but the differences between the 2 groups did not reach statistical significance. The 2 groups did not differ in terms of the occurrence of cholestasis during parenteral nutrition (63/121 vs 57/123; P = 0.444). Subgroup analysis of infants who had received more than three-quarters of their total daily fluid as parenteral nutrition showed, however, that the high manganese group developed significantly higher whole-blood manganese concentration [743.5 (498.0; 1211.0) vs 587.0 (438.0; 982.0); p = 0.037] and serum direct bilirubin concentration [84.0 (28.0; 170.0) vs 25.5 (9.0; 117.0); p < 0.001]. Although there was no significant difference in the occurrence of cholestasis (58/78 vs 49/82; p = 0.073), more infants in the high manganese group developed a more severe degree of direct hyperbilirubinaemia, with peak serum direct bilirubin > 100 mu mol/L (32/78 vs 20/82; p = 0.038). Conclusion: We conclude that the pathogenesis of parenteral nutrition-related cholestasis is probably multifactorial, and that high manganese intake is a significant contributory factor.

Fillebeen C, Ruchoux MM, Mitchell V, Vincent S, Benaissa M, Pierce A. 2001. Lactoferrin is synthesized by activated microglia in the human substantia nigra and its synthesis by the human microglial CHME cell line is upregulated by tumor necrosis factor alpha or 1-methyl-4-phenylpyridinium treatment. Molecular Brain Research 96(1-2):103-113.
Abstract: The presence of the iron-binding protein lactoferrin (Lf) in some specific areas of the central nervous system and particularly in the normal human substantia nigra, where it is found in dopaminergic (DA) neurons and some glial cells, led us to investigate Lf synthesis in this area. Lf mRNA were identified using in situ hybridization and found in small ameboid cells. These cells were identified using immunocytochemistry as activated microglia since they exhibited macrophage markers such as the CD68 and the CR1 antigens. Double immunofluorescent labeling confirmed that the two Lf immunostained cell populations were activated microglia and DA neurons. Since activated microglia contained both Lf and its messenger, these cells are the Lf producing cells. The presence of Lf in DA neurons in which no Lf messengers were visible, might be due to an endocytosis mechanism, DA neurons probably internalizing Lf produced in microglial cells located in their neighborhood. In neuropathological disorders, such as Alzheimer's and Parkinson's diseases, inflammatory process and oxidative stress are events that contribute to neuronal death. Since Lf concentration increases during these pathologies, we studied the level of Lf expression under these different stresses and showed, using RT-PCR, that the immortalized human embryonic microglial CHME cell line produced Lf transcripts under tumor necrosis factor a or 1-methyl-4-phenylpyridinium treatment whereas untreated cells did not. These data confirm that Lf is produced only when microglia are activated. (C) 2001 Elsevier Science B.V. All rights reserved.

Farin FM, Hitosis Y, Hallagan SE, Kushleika J, Woods JS, Janssen PS, Smith-Weller T, Franklin GM, Swanson PD, Checkoway H. 2001. Genetic polymorphisms of superoxide dismutase in Parkinson's disease. Mov Disord 16(4):705-707.
Abstract: Oxidative stress reactions may contribute to the pathogenesis of Parkinson's disease (PD). The superoxide dismutases potentially play significant roles in PD by detoxifying superoxide radical. We developed genomic DNA and cDNA-based sequencing assays to identify genetic variants in the copper/zinc superoxide dismutase (SOD1) and manganese superoxide dismutase (SOD2) genes. No genetic variants were detected in the gene encoding SOD 1 in DNA from 45 idiopathic PD cases and 49 controls from a population-based case-control study. However, we identified a previously described polymorphism of the mitochondrial targeting sequence consisting of a C47T in exon 2 of SOD2, which results in an alanine to valine substitution. We analyzed this SOD2 variant in DNA from 155 cases and 231 controls from the same study, using an allele-specific fluorogenic 5 ' nuclease assay, and found no differences in the distributions of allelic frequencies. These results indicate that SOD gene variants do not contribute to PD pathogenesis. (C) 2001 Movement Disorder Society.

Ebadi M, Govitrapong P, Sharma S, Muralikrishnan D, Shavali S, Pellett L, Schafer R, Albano C, Eken J. 2001. Ubiquinone (coenzyme Q(10)) and mitochondria in oxidative stress of Parkinson's disease. Biol Signals Recept 10(3-4): 224-253.
Abstract: Parkinson's disease is the second most common neurodegenerative disorder after Alzheimer's disease affecting approximately 1% of the population older than 50 years. There is a worldwide increase in disease prevalence due to the increasing age of human populations. A definitive neuropathological diagnosis of Parkinson's disease requires loss of dopaminergic neurons in the substantia nigra and related brain stem nuclei, and the presence of Lewy bodies in remaining nerve cells. The contribution of genetic factors to the pathogenesis of Parkinson's disease is increasingly being recognized. A point mutation which is sufficient to cause a rare autosomal dominant form of the disorder has been recently identified in the asynuclein gene on chromosome 4 in the much more common sporadic, or 'idiopathic' form of Parkinson's disease, and a defect of complex I of the mitochondrial respiratory chain was confirmed at the biochemical level. Disease specificity of this defect has been demonstrated for the parkinsonian substantia nigra. These findings and the observation that the neurotoxin 1-methyl-4-phenyl-1,2,3, 6-tetrahydropyridine (MPTP), which ca uses a Parkinson-like syndrome in humans, acts via inhibition of complex I have triggered research interest in the mitochondrial genetics of Parkinson's disease. Oxidative phosphorylation consists of five protein-lipid enzyme complexes located in the mitochondrial inner membrane that contain flavins (FMN, FAD), quinoid compounds (coenzyme Q(10), CoQ(10)) and transition metal compounds (iron-sulfur clusters, hemes, protein-bound copper). These enzymes are designated complex (NADH:ubiquinone oxidoreductase, EC 1.6. 5.3), complex II (succinate:ubiquinone oxidoreductase, EC 1.3.5.1), complex ill (ubiquinol:ferrocytochrome c oxidoreductase, EC 1.10.2.2), complex IV (ferrocytochrome c:oxygen oxidoreductase or cytochrome c oxidase, EC 1.9.3,1), and complex V (ATP syn thase, EC 3.6.1.34). A defect in mitochondrial oxidative phosphorylation, in terms of a reduction in the activity of NADH CoQ reductase (complex I) has been reported in the striatum of patients with Parkinson's disease, The reduction in the activity of complex I is found in the substantia nigra, but not in other areas of the brain, such as globus pallidus or cerebral cortex. Therefore, the specificity of mitochondrial impairment may play a role in the degeneration of nigrostriatal dopaminergic neurons. This view is supported by the fact that MPTP generating 1-methyl-4-phenylpyridine (MPP+) destroys dopaminergic neurons in the substantia nigra. Although the serum levels of CoQ(10) is normal in patients with Parkinson's disease, CoQ(10) is able to attenuate the MPTP-induced loss of striatal dopaminergic neurons. Copyright (C) 2001 S. Karger AG, Basel.

Duran E, Chacon JR. 2001. Parkinsonism probably induced by manganese. Rev Neurol 33(5):434-436.
Abstract: Introduction. In all cases of young persons with clinical Parkinson's disease it should be suspected that it is secondary to some primary disorder. Therefore a battery of diagnostic tests should be done before classification as idiopathic Parkinson's disease. Clinical case. A 31 year old woman whose only previous illness had been Graves disease. She complained of difficulty with movements of her right arm and leg for some months (she had problems with walking and with rapid, repeated movements of her right hand). She also complained of tremor of her right limbs at rest. She denied taking drugs, having dysphagia, dysarthria, visual changes or sphincter disorders. Neurological examination showed her to have monotonous speech, slight facial hypomimia, slight reduction in spontaneous blinking, walking with less swing of her right arm: postural tremor of both arms, worse on the right; bradykinesia (2/4) of both right limbs and rigidity (1/4), axial and of the right limbs. The results of all the investigations done to rule out secondary Parkinsonism were normal, except for the plasma manganese level which was raised, although it returned to normal when the probable source of exposure to this metal was removed. However, the alterations of movement only disappeared after treatment with levodopa was started. Conclusion. In cases of Parkinsonism in young adults secondary causes should always be rules out, such as exposure to certain metals.

Dobson J. 2001. Nanoscale biogenic iron oxides and neurodegenerative disease. FEBS Lett 496(1):1-5.
Abstract: One of the characteristics of many neurodegenerative diseases is the disruption of normal iron homeostasis in the brain. Recent experimental work indicates that nanoscale magnetic biominerals (primarily magnetite and maghemite) may be associated with senile plaques and tan filaments found in brain tissue affected by these diseases, These findings have important implications for our understanding of the role of iron in neurodegenerative disease as well as profound implications for their causes, In addition, the presence of biogenic magnetite in affected tissue should also provide improved mechanisms for early detection through the modification of MR] pulse sequences. (C) 2001 Federation of European Biochemical Societies. Published by Elsevier Science B.V. All rights reserved.

Datla KP, Blunt SB, Dexter DT. 2001. Chronic L-DOPA administration is not toxic to the remaining dopaminergic nigrostriatal neurons, but instead may promote their functional recovery, in rats with partial 6-OHDA or FeCl3 nigrostriatal lesions. Mov Disord 16(3):424-434.
Abstract: In this study, we have examined the effects of chronic L-3,4-dihydroxyphenylalanine (L-DOPA) administration on the remaining dopaminergic neurons in rats with 6-hydroxydopamine (6-OHDA) or buffered FeCl3 partial lesions to the nigrostriatal tract. L-DOPA administration increased the turnover of dopamine in the striatum. L-DOPA administration for 1 week produced an increase in the level of striatal RTI-121 binding, a specific marker for dopamine uptake sites: on the dopaminergic nerve terminals in the striatum. However, longer periods of L-DOPA treatment decreased the level of RTI-121 binding in the striatum. In the partial 6-OHDA lesion model, L-DOPA treatment had a time-dependent effect on the number of neurons demonstrating a dopaminergic phenotype i.e., neurons that are tyrosine hyrdoxylase (TH)immunopositive, on the lesioned side of the brain. In the first few weeks of treatment, L-DOPA decreased the number of TH-positive neurons but with long-term treatment, i.e., 24 weeks, L-DOPA increased the number of neurons demonstrating a dopaminergic phenotype, Even in the buffered FeCl3 infusion model, where the levels of iron were increased, L-DOPA treatment did not have any detrimental effects on the number of TH-positive neurons on the lesioned side of the brain. Consequently, chronic L-DOPA treatment does not have any detrimental effects to the remaining dopaminergic neurons in rats with partial lesions to the nigrostriatal tract, indeed in the 6-OHDA lesion model, long-term L-DOPA may increase the number of

neurons, demonstrating a dopaminergic phenotype. (C) 2001 Movement Disorder Society.

Dal-Pizzol F, Klamt F, Frota MLC, Andrades ME, Caregnato FF, Vianna MMR, Schroder N, Quevedo J, Izquierdo I, Archer T, Moreira JCF. 2001. Neonatal iron exposure induces oxidative stress in adult Wistar rat. Developmental Brain Research 130(1):109-114.
Abstract: Oxidative stress and excess of iron in the brain has been implicated in a variety of acute and chronic neurological conditions. The neonatal period is critical for the establishment of normal iron content in the adult brain. In the present study, the long-term oxidative effects of iron exposure during this period were assessed by treating Wistar rats orally with 0, 7.5 or 15 mg Fe+2/kg of body weight on postnatal days 10-12. Thiobarbituric acid reactive species, protein carbonyl, superoxide dismutase activity were measured at the age of 3 months. It was found that there was an increase in thiobarbituric acid reactive species and protein carbonyl in the substantia nigra of iron treated rats. In contrast, oxidative stress in the striatum was decreased. Superoxide dismutase activity was decreased in the substantia nigra iron treated rats. There were no differences in cerebellum measures among the groups. Our results demonstrated that iron supplementation in a critical neonatal period induced oxidative stress and modulated SOD activity in the adult life in selective brain regions. (C) 2001 Elsevier Science B.V. All rights reserved.

Curtis ARJ, Fey C, Morris CM, Bindoff LA, Ince PG, Chinnery PF, Coulthard A, Jackson MJ, Jackson AP, Mchale DP, Hay D, Barker WA, Markham AF, Bates D, Curtis A, Burn J. 2001. Mutation in the gene encoding ferritin light polypeptide causes dominant adult-onset basal ganglia disease. Nat Genet 28(4):350-354.
Abstract: We describe here a previously unknown, dominantly inherited, late-onset basal ganglia disease, variably presenting with extrapyramidal features similar to those of Huntington's disease (HD) or parkinsonism. We mapped the disorder, by linkage analysis, to 19q13.3, which contains the gene for ferritin light polypeptide (FTL). We found an adenine insertion at position 460-461 that is predicted to alter carboxy-terminal residues of the gene product. Brain histochemistry disclosed abnormal aggregates of ferritin and iron. Low serum ferritin levels also characterized patients. Ferritin, the main iron storage protein, is composed of 24 subunits of two types (heavy, H and light, L) which form a soluble, hollow sphere(1). Brain iron deposition increases normally with age, especially in the basal ganglia, and is a suspected causative factor in several neurodegenerative diseases (2) in which it correlates with visible pathology(3), possibly by its involvement in toxic free-radical reactions(4). We found the same mutation in five apparently unrelated subjects with similar extrapyramidal symptoms. An abnormality in ferritin strongly indicates a primary function for iron in the pathogenesis of this new disease, for which we propose the name 'neuroferritinopathy'.

Chun HS, Lee H, Son JH. 2001. Manganese induces endoplasmic reticulum (ER) stress and activates multiple caspases in nigral dopaminergic neuronal cells, SN4741. Neurosci Lett 316(1):5-8.
Abstract: Chronic exposure to manganese causes Parkinson's disease (PD)-like clinical symptoms (Neurotoxicology 5 (1984) 13; Arch. Neurol. 46 (1989) 1104; Neurology 56 (2001) 4). Occupational exposure to manganese is proposed as a risk factor in specific cases of idiopathic PD (Neurology 56 (2001) 8). We have investigated the mechanism of manganese neurotoxicity in nigral dopaminergic (DA) neurons using the DA cell line, SN4741 (J. Neurosci. 19 (1999) 10). Manganese treatment elicited endoplasmic reticulum (ER) stress responses, such as an increased level of the ER chaperone BiP, and simultaneously activated the ER resident caspase-12. Peak activation of other major initiator caspases-like activities, such as caspase-1, -8 and -9, ensued, resulting in activation of caspase-3-like activity during manganese-induced DA cell death. The neurotoxic cell death induced by manganese was significantly reduced in the Bcl-2-overexpressing DA cell lines. Our findings suggest that

manganese-induced neurotoxicity is mediated in part by ER stress and considerably ameliorated by Bcl-2 overexpression in DA cells. (C) 2001 Published by Elsevier Science Ireland Ltd.

Chun HS, Gibson GE, Degiorgio LA, Zhang H, Kidd VJ, Son JH. 2001. Dopaminergic cell death induced by MPP+, oxidant and specific neurotoxicants shares the common molecular mechanism. J Neurochem 76 (4):1010-1021.
Abstract: Recent etiological study in twins (Tanner et al. 1999) strongly suggests that environmental factors play an important role in typical, non-familial Parkinson's disease (PD), beginning after age 50. Epidemiological risk factor analyses of typical PD cases have identified several neurotoxicants, including MPP+ (the active metabolite of MPTP), paraquat, dieldrin, manganese and salsolinol. Here, we tested the hypothesis that these neurotoxic agents might induce cell death in our nigral dopaminergic cell line, SN4741 (Son et al 1999) through a common molecular mechanism. Our initial experiments revealed that treatment with both MPP + and the other PD-related neurotoxicants induced apoptotic cell death in SN4741 cells, following initial increases of H2O2-related ROS activity and subsequent activation of JNK1/2 MAP kinases. Moreover, we have demonstrated that during dopaminergic cell death cascades, MPP+, the neurotoxicants and an oxidant, H2O2 equally induce the ROS-dependent events. Remarkably, the oxidant treatment alone induced similar sequential molecular events: ROS increase, activation of JNK MAP kinases, activation of the PITSLRE kinase, p110, by both Caspase-1 and Caspase-8-like activities and apoptotic cell death. Pharmacological intervention using the combination of the antioxidant Trolox and a pan-caspase inhibitor Boc-(Asp)-fmk (BAF) exerted significant neuroprotection against ROS-induced dopaminergic cell death. Finally, the high throughput cDNA microarray screening using the current model identified downstream response genes, such as heme oxygenase-1, a constituent of Lewy bodies, that can be the useful biomarkers to monitor the pathological conditions of dopaminergic neurons under neurotoxic insult.

Chiueh CC. 2001. Iron overload, oxidative stress, and axonal dystrophy in brain disorders. Pediatr Neurol 25(2):138-147.
Abstract: Hallervorden-Spatz syndrome is an autosomal-recessive brain disorder with signs of extrapyramidal dysfunction and mental deterioration, which associate with iron accumulation in globus pallidus and substantia nigra pars reticulata. Studies of oxidant stress in parkinsonian animal models suggest a linkage of iron overload to axonal dystrophy. Redox cycling of iron complexes (i.e., ferrous citrate and hemoglobin) increases hydroxyl radicals, lipid peroxidation, axonal dystrophy, and necrotic or apoptotic cell death. An increase of oxidative stress in the basal ganglia because of redox cycling of iron complexes leads to dopamine overflow and psychomotor dysfunction. Iron overload-induced axonal dystrophy has been demonstrated consistently using in vitro and in vivo models with a prominent feature of lipid peroxidation. This iron-induced oxidative stress is often accentuated by ascorbate and oxidized glutathione, although it is suppressed by the following antioxidants: S-nitrosoglutathione or nitric oxide, MnSOD mimics, manganese, U-78517F, Trolox, and deferoxamine. Preconditioning induction of stress proteins (i.e., hemeoxygenase-1 and neuronal nitric oxide synthase) and hypothermia therapy suppress the generation of toxic reactive oxygen, lipid, and thiol species evoked by bioactive iron complexes in the brain. Finally, combined antioxidative therapeutics and gene induction procedures may prove to be useful for slowing progressive neurodegeneration caused by iron overload in the brain. (C) 2001 by Elsevier Science Inc. All rights reserved.

Chen JY, Tsao GC, Zhao QQ, Zheng W. 2001. Differential cytotoxicity of Mn(II) and Mn(III): Special reference mitochondrial [Fe-S] containing enzymes. Toxicol Appl Pharmacol 175(2):160-168.
Abstract: Manganese (Mn)-induced neurodegenerative toxicity has been associated with a distorted iron (Fe) metabolism at both systemic and cellular levels. In the current study, we examined whether the oxidation

states of Mn produced differential effects on certain mitochondrial [Fe-S] containing enzymes in vitro. When mitochondrial aconitase, which possesses a [4Fe-4S] cluster, was incubated with either Mn(II) or Mn(III), both Mn species inhibited the activities of aconitase. However, the IC10 (concentration to cause a 10% enzyme inhibition) for Mn(HI) was ninefold lower than that for Mn(II). Following exposure of mitochondrial fractions with Mn(II) or Mn(III), there was a significant inhibition by either Mn species in activities of Complex I whose active site contains five to eight [Fe-S] clusters. The dose-time response curves reveal that Mn(III) was more effective in blocking Complex I activity than Mn(II). Northern blotting was used to examine the expression of mRNAs encoding transferrin receptor (TfR), which is regulated by cytosolic aconitase. Treatment of cultured PC12 cells with Mn(II) and Mn(III) at 100 muM for 3 days resulted in 21 and 58% increases, respectively, in the expression of TfR mRNA. Further studies on cell growth dynamics after exposure to 25-50 muM Mn in culture media demonstrated that the cell numbers were much reduced in Mn(III)- treated groups compared to Mn(II)-treated groups, suggesting that Mn(III) is more effective than Mn(II) in cell killing. In cells exposed to Mn(II) and Mn(IH), mitochondrial DNA (mtDNA) was significantly decreased by 24 and 16%, respectively. In contrast, rotenone and MPP+ did not seem to alter mtDNA levels. These in vitro results suggest that Mn(IH) species appears to be more cytotoxic than Mn(II) species, possibly due to higher oxidative reactivity and closer radius resemblance to Fe. (C) 2001 Academic Press.

Chakraborty H, Ray SN, Chakrabarti S. 2001. Lipid peroxidation associated protein damage in rat brain crude synaptosomal fraction mediated by iron and ascorbate. Neurochem Int 39(4):311-317.
Abstract: In crude synaptosomal fractions from rat brain exposed to iron and ascorbate, enhanced lipid peroxidation (more than 3-fold compared to control), loss of protein thiols up to the extent of 40% compared to control, increased incorporation of carbonyl groups into proteins (more than 4.5-fold compared to control) and non-disulphide covalent cross-linking of membrane proteins have been observed. The phenomena are not inhibited by catalase or hydroxyl radical scavengers like mannitol or dimethyl sulphoxide. However. chain breaking antioxidants like alpha -tocopherol and butylated hydroxytoluene prevent both lipid peroxidation and accompanying protein oxidation. It is suggested that in this system lipid peroxidation propagated by the decomposition of preformed lipid hydroperoxides by iron and ascorbate is the primary event and products of the peroxidation process cause secondary protein damage. In view of high ascorbate content of brain and availability of several transition metals. such ascorbate mediated oxidative damage may be relevant in the aetiopathogenesis of several neurodegenerative disorders as well as ageing of brain. (C) 2001 Elsevier Science Ltd. All rights reserved.

Centonze D, Gubellini P, Bernardi G, Calabresi P. 2001. Impaired excitatory transmission in the striatum of rats chronically intoxicated with manganese. Exp Neurol 172(2):469-476.
Abstract: Chronic exposure to manganese (Mn) is known to produce a parkinsonian or dystonic state in humans caused by a rather selective involvement of the basal ganglia. Experimental observations suggest that secondary excitotoxic mechanisms play a crucial role in the development of Mn-induced neurodegeneration in the striatum, although the site of interference of Mn with glutamatergic transmission in this brain area is still unknown. To answer this question, in the present in vitro study, we investigated the physiological characteristics of striatal excitatory synaptic transmission in a rat model of Mn intoxication. We found that chronic Mn greatly increased both frequency and amplitude of spontaneous excitatory postsynaptic potentials, in the absence of appreciable changes of intrinsic membrane properties of striatal cells. The sensitivity of striatal neurons to glutamate AMPA and NMDA receptor stimulation was unaffected by Mn poisoning, as demonstrated by comparing the membrane responses produced in control and treated rats to the application of selective agonists of these receptors and to the direct activation of corticostriatal

glutamatergic fibers. In addition, also paired-pulse facilitation was unaltered by Mn treatment, indicating that this toxin does not affect the pre- and postsynaptic mechanisms responsible for the appearance of this short-term form of synaptic plasticity at corticostriatal synapses. It is concluded, therefore, that hyperactivity of corticostriatal neurons, rather than increased postsynaptic sensitivity to glutamate, accounts for the abnormal excitation of striatal neurons in the course of Mn intoxication. (C) 2001 Elsevier Science.

Carpenter DO. 2001. Effects of metals on the nervous system of humans and animals. Int J Occup Med Environ Health 14(3):209-18.
Abstract: Several metals have toxic actions on nerve cells and neurobehavorial functioning. These toxic actions can be expressed either as developmental effects or as an increased risk of neurodegenerative diseases in old age. The major metals causing neurobehavioral effects after developmental exposure are lead and methylmercury. Lead exposure in young children results in a permanent loss of IQ of approximately 5 to 7 IQ points, and also results in a shortened attention span and expression of anti-social behaviors. There is a critical time period (<2 years of age) for development of these effects, after which the effects do not appear to be reversible even if blood lead levels are lowered with chelation. Methylmercury has also been found to have effects on cognition at low doses, and prenatal exposure at higher levels can disrupt brain development. Metals have also been implicated in neurodegenerative diseases, although it is unlikely that they are the sole cause for any of them. Elevated aluminum levels in blood, usually resulting from kidney dialysis at home with well water containing high aluminum, result in dementia that is similar to but probably different from that of Alzheimer's disease. However, there is some epidemiological evidence for elevated risk of Alzheimer's in areas where there is high concentration of aluminum in drinking water. Other metals, especially lead, mercury, manganese and copper, have been implicated in amvotrophic lateral sclerosis and Parkinson's disease.

Calabresi P, Ammassari-Teule M, Gubellini P, Sancesario G, Morello M, Centonze D, Marfia GA, Saulle E, Passino E, Picconi B, Bernardi G. 2001. A synaptic mechanism underlying the behavioral abnormalities induced by manganese intoxication. Neurobiol Dis 8(3):419-432.
Abstract: In the present study we have characterized a rad: model of manganese (Mn) intoxication leading to behavioral disinhibition in the absence of major motor alterations. These behavioral changes were associated with significantly increased brain Mn levels but were uncoupled to anatomical lesions of the striatum or to morphological and cytochemical changes of the nigrostriatal dopaminergic pathway. The analysis of this model at cellular level showed an enhanced dopaminergic inhibitory control of the corticostriatal excitatory transmission via presynaptic DP-like dopamine (DA) receptors in slices obtained from Mn-treated rats. Conversely, the use of agonists acting on presynaptic purinergic, muscarinic, and glutamatergic metabotropic receptors revealed a normal sensitivity. Moreover, membrane responses recorded from single dopaminergic neurons following activation of D2 DA eutoreceptors were also unchanged following Mn intoxication. Thus, our findings indicate a selective involvement of the DP-like DA receptors located on glutamatergic corticostriatal terminals in this pathological condition and suggest that the behavioral symptoms described in the "early" clinical phase of manganism may be caused by an abnormal dopaminergic inhibitory control on corticostriatal inputs. The identification of the synaptic mechanism underlying the "early" phase of Mn intoxication might have a critical importance to understand the causes of the progression of this pathological condition towards an "established" phase characterized by motor abnormalities and anatomical lesions of the basal ganglia. (C) 2001 Academic Press.

Bush AI, Goldstein LE. 2001. Specific metal-catalysed protein oxidation reactions in chronic degenerative disorders of ageing: focus on Alzheimer's disease

and age-related cataracts. Novartis Found Symp 235:26-38; discussion 38-43.
Abstract: Abnormalities of protein aggregation and deposition may play an important role in the pathophysiology of a diverse set of chronically progressive degenerative disorders including Alzheimer's disease, amyotrophic lateral sclerosis, Parkinson's disease and age-related cataracts. We propose that aberrant metalloprotein reactions may be a common denominator in these diseases. In these instances, an abnormal reaction between a protein and redox active metal ions (especially copper or iron) promotes the generation of reactive oxygen species, and possibly, protein radicalization. These products then lead to chemical modification of the protein, alterations in protein structure and solubility, and oxidative damage to surrounding tissue. In this review, we explore these ideas by focusing on two common diseases of ageing, Alzheimer's disease and age-related cataracts. Understanding the metalloprotein biochemistry in both diseases may lead to a better understanding of the underlying pathophysiology in both disorders and suggest novel targets for therapeutic agents.

Burn DJ, Jaros E. 2001. Multiple system atrophy: cellular and molecular pathology. Journal of Clinical Pathology-Molecular Pathology 54(6): 419-426.
Abstract: Multiple system atrophy is an adult onset neurodegenerative disease, featuring parkinsonism, ataxia, and autonomic failure, in any combination. The condition is relentlessly progressive and responds poorly to treatment. Death occurs on average six to seven years after the onset of symptoms. No familial cases of multiple system atrophy have been reported, and no environmental factors have been robustly implicated as aetiological factors. However, analytical epidemiological studies are hampered because the condition is relatively rare. The discovery of the gliai cytoplasmic inclusion (GCI) in 1989 helped to define multiple system atrophy as a clinicopathological entity, and drew attention to the prominent, if not primary, role played by the oligodendrocyte in the pathogenesis of the condition. Subsequently, GCIs were shown to be positive for alpha -synuclein, with immunostaining for this protein indicating that white matter pathology was more widespread than had previously been recognised. The presence of alpha -synuclein in GCIs provides a link with Parkinson's disease, dementia with Lewy bodies, and neurodegeneration with brain iron accumulation, type 1 (or Hallervorden-Spatz syndrome), in which a-synuclein is also found within Lewy bodies. This has led to the term "synucleinopathy" to embrace this group of conditions. The GCIs of multiple system atrophy contain a range of other cytoskeletal proteins. It is unknown how fibrillogenesis occurs, and whether there is primary oligodendrocytic dysfunction, which then disrupts the neurone/axon as a consequence of the glial pathology, or whether the oligodendrocytic changes merely represent an epiphenomenon. Further research into this devastating condition is urgently needed to improve our understanding of the pathogenesis, and also to produce new treatment approaches.

Burdo JR, Menzies SL, Simpson IA, Garrick LM, Garrick MD, Dolan KG, Haile DJ, Beard JL, Connor JR. 2001. Distribution of divalent metal transporter 1 and metal transport protein 1 in the normal and Belgrade rat. J Neurosci Res 66 (6): 1198-1207.
Abstract: Iron accumulation in the brain occurs in a number of neurodegenerative diseases. Two new iron transport proteins have been identified that may help elucidate the mechanism of abnormal iron accumulation. The Divalent Metal Transporter 1 (DMT1), is responsible for iron uptake from the gut and transport from endosomes. The Metal Transport Protein 1 (MTP1) promotes iron export. In this study we determined the cellular and regional expression of these two transporters in the brains of normal adult and Belgrade rats. Belgrade rats have a defect in DMT1 that is associated with lower levels of iron in the brain. In the normal rat, DMT1 expression is highest in neurons in the striatum, cerebellum, thalamus, ependymal cells lining the third ventricle, and

vascular cells throughout the brain. The staining in the ependymal cells and endothelial cells suggests that DMT1 has an important role in iron transport into the brain. In Belgrade rats, there is generalized decrease in immunodetectable DMT1 compared to normal rats except in the ependymal cells. This decrease in immunoreactivity, however, was absent on immunoblots. The immunoblot analysis indicates that this protein did not upregulate to compensate for the chronic defect in iron transport. MTP1 staining is found in most brain regions. MTP1 expression in the brain is robust in pyramidal neurons of the cerebral cortex but is not detected in the vascular endothelial cells and ependymal cells. MTP1 staining in Belgrade rats was decreased compared to normal, but similar to DMT1 this decrease was not corroborated by immunoblotting. These results indicate that DMT1 and MT are involved in brain iron transport and this involvement is regionally and cellularly specific. (C) 2001 Wiley-Liss, Inc.

Brooks DG, Lech G, Vandenborne K, Wilson RB, Stambolian D, Chance B, Kelley RI. 2001. A novel, autosomal-recessive form of cardioskeletal myopathy and neutropenia with iron deficiency and Parkinsonism. Am J Hum Genet 69(4):479.

Boruchowska M, Lankosz M, Adamek D, Korman A . 2001. PIXE analysis of human brain tissue. X-Ray Spectrometry 30(3):174-179.
Abstract: The proton induced x-ray emission (PIXE) technique was used for localized analysis of trace elements in small quantities of medical samples. Both white and gray matter of the human brain were studied. The tissue samples were taken at autopsy from patients aged from 17 to 88 years. Qualitative and quantitative analyses were performed. NIST SRM 1577b Bovine Liver and SRM 1566a Oyster Tissue were used as external standards for the determination of trace element mass fractions in the brain specimens. The main goal of these preliminary studies was to evaluate approaches for the quantitative analysis of medical materials using PIXE taking into account the effect of patient age on the accumulation of elements and the regional distribution of elements in human brain. Detection and quantification limits were calculated for selected elements. Elevation of Zn concentration with age of patients was observed for white mater of the human brain. Elements such as S, K, Ca, Mn, Fe, Cu and Zn clearly exhibit higher mass fractions in grey matter than in white matter in all cases. This investigation confirms that PIXE is a useful technique for multielement analysis of tissue samples with good sensitivity. Copyright (C) 2001 John Wiley & Sons, Ltd.

Blanchard-Fillion B, Souza JM, Friel T, Jiang GCT, Vrana K, Sharov V, Barron L, Schoneich C, Quijano C, Alvarez B, Radi R, Przedborski S, Fernando GS, Horwitz J, Ischiropoulos H. 2001. Nitration and inactivation of tyrosine hydroxylase by peroxynitrite. J Biol Chem 276(49):46017-46023.
Abstract: Tyrosine hydroxylase (TH) is modified by nitration after exposure of mice to 1-methyl-4-phenyl-1,2,3,6-tetrahydrophenylpyridine. The temporal association of tyrosine nitration with inactivation of TH activity in vitro suggests that this covalent post-translational modification is responsible for the in vivo loss of TH function (Ara, J., Przedborski, S., Naini, A. B., Jackson-Lewis, V., Trifiletti, R. R., Horwitz, J., and Ischiropoulos, H. (1998) Proc. Natl. Acad Sci. U. S. A. 95, 7659 -7663). Recent data showed that cysteine oxidation rather than tyrosine nitration is responsible for TH inactivation after peroxynitrite exposure in vitro (Kuhn, D. M., Aretha, C. W., and Geddes, T. J. (1999) J. Neurosci. 19, 10289-10294). However, re-examination of the reaction of peroxynitrite with purified TH failed to produce cysteine oxidation but resulted in a concentration-dependent increase in tyrosine nitration and inactivation. Cysteine oxidation is only observed after partial unfolding of the protein. Tyrosine residue 423 and to lesser extent tyrosine residues 428 and 432 are modified by nitration. Mutation of Tyr(423) to Phe resulted in decreased nitration as compared with wild type protein without loss of activity. Stopped-flow experiments reveal a second order rate constant of (3.8 +/- 0.9) X 10(3) M-1 s(-1) at pH 7.4 and 25 degreesC for the reaction of peroxynitrite with TH. Collectively, the data indicate that peroxynitrite

reacts with the metal center of the protein and results primarily in the nitration of tyrosine residue 423, which is responsible for the inactivation of TH.

Bidmon HJ, Emde B, Oermann E, Kubitz R, Witte OW, Zilles K. 2001. Heme oxygenase-1 (HSP-32) and heme oxygenase-2 induction in neurons and glial cells of cerebral regions and its relation to iron accumulation after focal cortical photothrombosis. Exp Neurol 168(1):1-22.
Abstract: Cerebral ischemic injury results in the liberation of heme from degenerating heme-containing proteins. The neurotoxic heme is usually detoxified by the constitutive heme oxygenase-a (HO-2) and its inducible isoform HO-l(heat shock protein 32) resulting in the formation of biliverdin which becomes reduced to bilirubin, carbon monoxide (CO), and iron. Biliverdin and bilirubin have antioxidative properties whereas CO is discussed as a signaling molecule, Iron if it remains free could catalyze Haber-Weiss and Fenton reactions causing the formation of highly toxic radicals, We have studied the alterations of cerebral HO-2 and HO-1 in relation to iron accumulations after defined cortical photothrombosis within the hindlimb area of the rat. HO-2 immunohistochemistry showed that the number of HO-2-positive neurons in most perilesional regions remained constant. However, much stronger systemic immunoreactivity for HO-2 was observed between days 1 and 7 postlesion, For HO-1 a systemic increase of immunoreactivity occurred also between days 1 and 7, In addition HO-1-positive astrocytes and microglia appeared as early as 4 h postlesion and increased up to day 3 followed by a sharp decline toward day 14 within the injured hemisphere. HO-1-positive astrocytes and microglia occurred in ipsilateral cortex, corpus callosum, hippocampus, striatum, and thalamic nuclei, Additionally an increase of HO-1 in myelin-associated globulin-positive oligodendrocytes was found in ipsilateral and contralateral cortex, Next to the lesion iron accumulation occurred after day 3 and increased strongly toward day 14 at times when HO-1 and -2 had decreased, suggesting that HO activity does not directly contribute to postlesional iron deposition. (C) 2000 Academic Press.

Berg D, Gerlach M, Youdim MBH, Double KL, Zecca L, Riederer P, Becker G. 2001. Brain iron pathways and their relevance to Parkinson's disease. J Neurochem 79(2):225-236.
Abstract: A central role of iron in the pathogenesis of Parkinson's disease (PD), due to its increase in substantia nigra pars compacta dopaminergic neurons and reactive microglia and its capacity to enhance production of toxic reactive oxygen radicals, has been discussed for many years. Recent transcranial ultrasound findings and the observation of the ability of iron to induce aggregation and toxicity of alpha -synuclein have reinforced the critical role of iron in the pathogenesis of nigrostriatal injury. Presently the mechanisms involved in the disturbances of iron metabolism in PD remain obscure. In this review we summarize evidence from recent studies suggesting disturbances of iron metabolism in PD at possibly different levels including iron uptake, storage, intracellular metabolism, release and post-transcriptional control. Moreover we outline that the interaction of iron with other molecules, especially a-synuclein, may contribute to the process of neurodegeneration. Because many neurodegenerative diseases show increased accumulation of iron at the site of neurodegeneration, it is believed that maintenance of cellular iron homeostasis is crucial for the viability of neurons.

Becker G, Berg D. 2001. Neuroimaging in basal ganglia disorders: Perspectives for transcranial ultrasound. Mov Disord 16(1):23-32.
Abstract: Transcranial sonography is a new diagnostic tool. allowing nor only the evaluation of cerebral arteries but also the two-dimensional display of the brain parenchyma, In this review We Will summarize basics of the application, the ultrasound anatomy of the brain and sonographic findings in some movement disorders. While in normal adults basal ganglia nuclei are hypoechogenic, they are hyperechogenic in certain basal ganglia disorders. In Parkinson's disease, for example, the substantia nigra can be depicted as a distinctly echogenic area, An elevated echogenicity of the

lentiform nuclei was noticed in patients with primary adult-onset dystonia. In both disorders the altered echogenicity may arise from higher heavy metal tissue content (i.e. iron in Parkinson's disease and copper in primary dystonia). Our findings converge to the hypothesis that transcranial ultrasound sensitively detects pathological metal accumulation not identified by other neuroimaging techniques (CT and MRI) and therefore provides new insights in the diagnosis of basal ganglia disorders. The implications of these findings for the understanding of the pathogenesis and its usefulness for the early diagnosis of movement disorders are outlined. Mov. Disord. 16:33-32, 2001. (C) 2001 Movement Disorder Society.

Barzilai A, Melamed E, Shirvan A. 2001. Is there a rationale for neuroprotection against dopamine toxicity in Parkinson's disease? Cell Mol Neurobiol 21(3): 215-235.
Abstract: Parkinson's disease is a progressive neurological disease caused by rather selective degeneration of the dopaminergic neurons in the substantia nigra. Though subject to intensive research, the etiology of this nigral loss is still undetermined and treatment is basically symptomatic. The current major hypothesis is that nigral neuronal death in PD is due to excessive oxidative stress generated by auto and enzymatic oxidation of the endogenous neurotransmitter dopamine (DA), the formation of neuromelanin (MM) and the presence of a high concentration of iron. In this review article although we concisely describe the effects of NM and iron on neuronal survival, we mainly focus on the molecular mechanisms of DA-induced apoptosis, DA exerts its toxic effects through its oxidative metabolites either in vitro or in vivo The oxidative metabolites then activate a very intricate web of signals, which culminate in cell death. The signal transduction pathways and genes, which are associated with DA toxicity are described in detail.

Barthel H, Sorger D, Kuhn HJ, Wagner A, Kluge R, Hermann W. 2001. Differential alteration of the nigrostriatal dopaminergic system in Wilson's disease investigated with [I-123]beta-CIT and high-resolution SPET. Eur J Nucl Med 28(11):1656-1663.
Abstract: Wilson's disease (WD) is a copper deposition disorder which can result in a number of extrapyramidal motoric symptoms such as parkinsonism. Therefore, this study was carried out to investigate, for the first time, nigrostriatal dopaminergic function in WD in relation to different courses and severity of the disease. Using high-resolution single-photon emission tomography (SPET) after administration of 2 beta -carbomethoxy-3 beta-(4[I-123]iodophenyl)tropane ([I-123]beta -CIT), striatal dopamine transporters (DAT) were imaged in 43 WD patients and a control group of ten subjects. From the SPET images, specific [I-123]beta -CIT binding ratios were obtained for the caudate heads, putamina and entire corpus striatum. In addition, to evaluate a putative dissociation between the caudate and putaminal [123I]beta -CIT binding ratios, the ratio between these binding ratios was calculated (CA/PU ratio). The SPET data were compared with clinical data on the course of the disease (CD), the severity of neurological symptoms and the degree of hepatic alteration. Whereas the specific regional [123I]beta -CIT binding ratios in patients with asymptomatic/hepatic CD did not differ from those in the control group (e.g. striatal ratios: 13.4 +/-3.0 vs 11.7 +/-2.8), in patients with neurological CD the ratios were significantly reduced for all striatal substructures (P=0.003 after one-factor ANOVA). For the different subgroups a tendency was detected towards a stepwise decrease in the specific [I-123]beta -CIT binding ratios from pseudo-sclerosis CD (9.4 +/-2.3), through pseudo-parkinsonian CD (9.1 +/-2.1) to arrhythmic-hyperkinetic CD (8.5 +/-1.6). However, these group differences reached significance only for the comparison with asymptomatic/hepatic CD (P=0.02). The CA/PU ratio was significantly higher in WD than in the control group (1.30 +/-0.19 vs 1.11 +/-0.08; P=0.003). Severity of neurological symptoms was significantly correlated with all specific regional [I-123]beta -CIT binding ratios (r=-0.49 to -0.57). For degree of liver alteration, significant correlations were obtained with the putaminal binding ratio

(r=-0.37) and the CA/PU ratio (r=0.44). From these results is concluded that in WD the nigrostriatal dopaminergic function is compromised to varying extents. The degree of this presynaptic alteration of dopaminergic neurotransmission depends on the clinical course and severity of this copper deposition brain disorder and also varies in the different striatal substructures.

Barreto WJ, Barreto SRG, Santos MA, Schimidt R, Paschoal FMM, Mangrich AS, Deoliveira LFC. 2001. Interruption of the MnO2 oxidative process on dopamine and L-dopa by the action of S2O32-. J Inorg Biochem 84(1-2): 89-96.
Abstract: The oxidation effects of Mn2+, Mn3- or MnO2 on dopamine can be studied in vitro and, therefore, this offers a model of the auto-oxidation process that appears naturally in neurons causing Parkinsons disease. The use of MnO2 as an oxidizer in aqueous solution at pH 7 causes the oxidation of catecholamines (L-dopa, dopamine, noradrenaline and adrenaline) to melanin, However, this work shows that, in water at pH 6-7. the oxidation of catecholamines by MnO2 in the presence of sodium thiosulphate (Na2S2O3) occurs by other mechanisms. For dopamine and L-dopa, MLCT complexes were formed with bands at 312, 350 (sh), 554 (sh) nm, and an intense band at 597 nm (epsilon congruent to 4 X 10(3) M-1 cm(-1)) and at ca. 336, 557 (sh) nm, and an intense band at 597 nm (epsilon approximate to 6 X 10(3) M-1 cm(-1)), respectively. The latter transitions were assigned to d(pi)-->pi*-SQ. Noradrenaline and adrenaline do not form this blue complex in solution, but generate soluble oxidized compounds. The resonance Raman spectra of these complexes in solution showed bands at 950, 1006, 1258, 1378, 1508 and 1603 cm(-1) for the complex derivation of L-dopa and at 948, 1010, 1255, 1373, 1510 and 1603 cm(-1) for the dopamine-derived compound. The most intense Raman band at ca. 1378 cm(-1) was assigned to C-O stretching with major C-1-C-2 characteristics and indicated that dopamine and L-dopa do not occur complexed with manganese in the catecholate or quinone form, but suggests an intermediate compound such as an anionic o-semiquinone (SQ(-)), forming a complex such as [Mn(II)(SQ(-))(3)](-). All enhanced Raman frequencies are characteristic of the benzenic ring without the participation of the aminic nitrogen. A mechanism is proposed for the Formation of the dopamine and L-dopa complexes and a computational simulation was performed to support it. (C) 2001 Elsevier Science B.V. All rights reserved.

Armstrong C, Leong W, Less GJ. 2001. Comparative effects of metal chelating agents on the neuronal cytotoxicity induced by copper (Cu+2), iron (Fe+3) and zinc in the hippocampus. Brain Res 892(1):51-62.
Abstract: The ability of metal chelating agents to prevent neuronal death caused by intra-hippocampal injections of cupric sulphate, ferric citrate and zinc chloride was investigated. Ammonium tetrathiomolybdate was itself toxic after injection into the hippocampus, but this toxicity was reduced by formation of a metal ion/tetrathiomolybdate complex with Cu2+. Disodium bathocuproine disulphonate (BCDS) prevented neuronal death caused by Cu+2, but not that induced by Fe+3 or Zn+2. Desferrioxamine prevented death caused by Fe+3, had no significant effect of the toxicity of Zn+2 and increased that caused by Cu+2. Even though N,N,N',N'-tetrakis(2-pyridylmethyl)ethylenediamine (TPEN) has a higher affinity for Cu+2 than for Zn+2, TPEN had no effect on the toxicity of Cu+2 while totally preventing damage caused by Fe+3 or Zn+2. Ethylenediaminetetra-acetic acid (EDTA) prevented the toxicity of all three metal ions. Motor seizure activity occurred in most rats after injections of Fe+3; or combinations of Cu+2 plus TPEN, or 4 nmol Fe+3 plus 0.1 nmol desferrioxamine. However, apart from the low dose desferrioxamine/Fe+3 combination, only the occasional brain contained seizure-induced neuronal loss in limbic regions outside the injected hippocampus, and these brains were not used for analysis. Seizure activity was found even with very low levels of Cu+2 with a fixed amount of TPEN (a ratio of Cu+2/TPEN of 1:100), but the extent of hippocampal damage in these brains was not significantly different to that caused by injections of saline. These studies demonstrate that idiosyncratic interactions can occur between metal ions and chelating agents. Thus

further investigations are needed before chelating agents can be examined for their protective properties in various neurodegenerative diseases. (C) 2001 Elsevier Science B.V. All rights reserved.

Andreassen OA, Ferrante RJ, Dedeoglu A, Albers DW, Klivenyi P, Carlson EJ, Epstein CJ, Beal MF. 2001. Mice with a partial deficiency of manganese superoxide dismutase show increased vulnerability to the mitochondrial toxins malonate, 3-nitropropionic acid, and MPTP. Exp Neurol 167(1): 189-195.
Abstract: There is substantial evidence implicating mitochondrial dysfunction and free radical generation as major mechanisms of neuronal death in neurodegenerative diseases. The major free radical scavenging enzyme in mitochondria is manganese superoxide dismutase (SOD2). In the present study we investigated the susceptibility of mice with a partial deficiency of SOD2 to the neurotoxins 1-methyl-4-phenyl-1,2,5,6-tetrahydro-pyridine (MPTP), 3-nitropropionic acid (3-NP), and malonate, which are commonly used animal models of Parkinson's and Huntington's disease, Heterozygous SOD2 knockout (SOD2(+/-)) mice showed no evidence of neuropathological or behavioral abnormalities at 2-4 months of age. Compared to littermate wild-type mice, mice with partial SOD2 deficiency showed increased vulnerability to dopamine depletion after systemic MPTP treatment and significantly larger striatal lesions produced by both 3-NP and malonate. SOD2(+/-) mice also showed an increased production of "hydroxyl" radicals after malonate injection measured with the salicylate hydroxyl radical trapping method. These results provide further evidence that reactive oxygen species play an important role in the neurotoxicity of MPTP, malonate, and 3-NP. These findings show that a subclinical deficiency in a free radical scavenging enzyme may act in concert with environmental toxins to produce selective neurodegeneration. (C) 2001 Academic Press.

Andersen JK. 2001. Do alterations in glutathione and iron levels contribute to pathology associated with Parkinson's disease? Novartis Found Symp 235:11-20; discussion 20-5 .
Abstract: A growing body of evidence has implicated oxidative stress as an important factor in the neuropathology associated with Parkinson's disease. Dopaminergic nigrostriatal neurons, the predominant cells lost in Parkinson's, are believed to be highly prone to oxidative damage due to the propensity for dopamine to auto-oxidize and thereby produce elevated levels of hydrogen peroxide and catecholamine quinones. Hydrogen peroxide formed during this process can either be converted by iron to form highly reactive hydroxyl radicals or removed through reduction by glutathione. Glutathione can also conjugate with quinones formed during dopamine oxidation preventing them from facilitating the release of iron from the iron-storage molecule ferritin. Alterations in both iron and glutathione levels in the substantia nigra have been correlated with the neuronal degeneration accompanying Parkinson's disease but a direct causative role for either has yet to be definitively proved. We will discuss the use of genetically engineered cell and mouse lines generated in our laboratory as models to examine the role that alterations in iron and glutathione levels may play in neurodegeneration of dopaminergic neurons of the substantia nigra associated with Parkinson's disease, and how these two parameters may interact with one another to bring this about.

Alcaraz-Zubeldia M, Rojas P, Boll C, Rios C. 2001. Neuroprotective effect of acute and chronic administration of copper (II) sulfate against MPP+ neurotoxicity in mice. Neurochem Res 26(1):59-64.
Abstract: Neurodegenerative effects of MPP+, the main metabolite of MPTP include dopamine (DA) depletion and enhanced lipid peroxidation (LPO) in mice striata, both associated to free radicals overproduction. Since copper is related to several antioxidant enzymes, we tested its neuroprotective effect against MPP+-induced neurotoxicity (20 mug/3 mul). CuSO4 pretreatment was administrated by either acute (2.5 mg/kg, i.p) or chronic (350 or 700 mg/l doses through drinking water, for 30 days) schemes. Acute administration blocked MPP+-induced striatal LPO only when

administered 16 or 24 hours before MPP+, and prevented the DA-depleting effect only at 24 hours. Chronic CuSO4 prevented the LPO increase, and blocked the DA depletion only at the higher dose used (700 mg/l). Neuroprotective effect of CuSO4 was dependent on the dose and the time of pretreatment, which suggest that this lag could be related with mechanisms of activation or synthesis of copper-dependent proteins responsible of cellular defense against MPP+.

Alcaraz-Zubeldia M, Montes S, Rios C. 2001. Participation of manganese-superoxide dismutase in the neuroprotection exerted by copper sulfate against 1-methyl 4-phenylpyridinium neurotoxicity. Brain Res Bull 55 (2): 277-279.
Abstract: Neurodegenerative effects of 1-methyl-4-phenylpyridinium (MPP +), the main metabolite of the neurotoxin 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) include enhancement of lipid peroxidation in the striatum of mice, associated to overproduction of free radicals. Copper acts as a prosthetic group of several copper-dependent antioxidant enzymes, and we previously showed the neuroprotective effect of CuSO4 pretreatment against the MPP+-induced neurotoxicity. In those studies, acute administration of CuSO4 (2.5 mg/kg) blocked MPP+-induced striatal lipid peroxidation, suggesting the activation of Cu-dependent proteins that defend neurons from damage elicited by free radicals. In the present study, we evaluated the activity of superoxide dismutase in mice pretreated with CuSO4 16 h or 24 h prior to MPP+ administration. Copper administration produced a specific and significant increase in manganese superoxide dismutase activity in both the CuSO4/saline (fivefold increase) and the CuSO4/MPP+ groups of animals (sevenfold increase). The Na2SO4/MPP+ group showed a twofold increase in manganese superoxide dismutase activity versus control levels. The results suggest that the load of copper activating manganese-dependent superoxide dismutase could be responsible for neuroprotection against the MPP+ insult. (C) 2001 Elsevier Science Inc.

Kim TD, Paik SR, Yang CH, Kim J. 2000 Dec. Structural changes in alpha-synuclein affect its chaperone-like activity in vitro . Protein Sci 9(12): 2489-96.
Abstract: Alpha-synuclein, a major constituent of Lewy bodies (LBs) in Parkinson's disease (PD), has been implicated to play a critical role in synaptic events, such as neuronal plasticity during development, learning, and degeneration under pathological conditions, although the physiological function of alpha-synuclein has not yet been established. We here present biochemical evidence that recombinant alpha-synuclein has a chaperone-like function against thermal and chemical stress in vitro. In our experiments, alpha-synuclein protected glutathione S-transferase (GST) and aldolase from heat-induced precipitation, and alpha-lactalbumin and bovine serum albumin from dithiothreitol (DTT)-induced precipitation like other molecular chaperones. Moreover, preheating of alpha-synuclein, which is believed to reorganize the molecular surface of alpha-synuclein, increased the chaperone-like activity. Interestingly, in organic solvents, which promotes the formation of secondary structure, alpha-synuclein aggregated more easily than in its native condition, which eventually might abrogate the chaperone-like function of the protein. In addition, alpha-synuclein was also rapidly and significantly precipitated by heat in the presence of Zn2+ in vitro, whereas it was not affected by the presence of Ca2+ or Mg2+. Circular dichroism spectra confirmed that alpha-synuclein underwent conformational change in the presence of Zn2+. Taken together, our data suggest that alpha-synuclein could act as a molecular chaperone, and that the conformational change of the alpha-synuclein could explain the aggregation kinetics of alpha-synuclein, which may be related to the abolishment of the chaperonic-like activity.

Kidd PM. 2000 Dec. Parkinson's disease as multifactorial oxidative neurodegeneration: implications for integrative management. Altern Med Rev 5(6):502-29.
Abstract: Parkinson's disease (PD) is the most common movement

pathology, severely afflicting dopaminergic neurons within the substantia nigra (SN) along with non-dopaminergic, extra-nigral projection bundles that control circuits for sensory, associative, premotor, and motor pathways. Clinical, experimental, microanatomic, and biochemical evidence suggests PD involves multifactorial, oxidative neurodegeneration, and that levodopa therapy adds to the oxidative burden. The SN is uniquely vulnerable to oxidative damage, having high content of oxidizable dopamine, neuromelanin, polyunsaturated fatty acids, and iron, and relatively low antioxidant complement with high metabolic rate. Oxidative phosphorylation abnormalities impair energetics in the SN mitochondria, also intensifying oxygen free radical generation. These pro-oxidative factors combine within the SN dopaminergic neurons to create extreme vulnerability to oxidative challenge. Epidemiologic studies and long-term tracking of victims of MPTP (1-methyl-4-phenyl-1,2,3,6,-tetrahydropyridine) poisoning, suggest oxidative stress compounded by exogenous toxins may trigger the neurodegenerative progression of PD. Rational, integrative management of PD requires: (1) dietary revision, especially to lower calories; (2) rebalancing of essential fatty acid intake away from pro-inflammatory and toward anti-inflammatory prostaglandins; (3) aggressive repletion of glutathione and other nutrient antioxidants and cofactors; (4) energy nutrients acetyl L-carnitine, coenzyme Q10, NADH, and the membrane phospholipid phosphatidylserine (PS), (5) chelation as necessary for heavy metals; and (6) liver P450 detoxification support.

Sato K, Ueyama H, Arakawa R, Kumamoto T, Tsuda T. 2000 Nov. [A case of welder presenting with parkinsonism after chronic manganese exposure]. Rinsho Shinkeigaku 40(11):1110-5.
Abstract: A 56-year-old welder working for 30 years developed postural instability and writing clumsiness since October, 1998. Neurologic findings revealed dystonia of the bilateral shoulders and distal four limbs as well as parkinsonism such as masked face, bradykinesia, rigidity, and retropulsion. Brain MRI showed hyperintensity lesions on T1-weighted images in the bilateral globus pallidus, midbrain, pontine tegmentum, dentate nucleus and cerebral white matter, which reduced in size and density after ten months. The diagnosis of manganese poisoning was made by the high manganese levels of both serum and urine, and by the marked elevated urinary manganese level after administration of the cheleting agent. We pointed out the diagnostic significance of brain MRI in patients with chronic manganese exposure.

Kuzuhara S. 2000 Oct. [Essential points to differentiate various diseases causing parkinsonism]. Nippon Rinsho 58(10):2049-53.
Abstract: This review article deals with the cardinal features to differentiate various conditions which present with parkinsonism, other than Parkinson's disease, Lewy body disease, progressive supranuclear palsy and corticobasal degeneration. Special attention is paid to the distinctive clinical features, laboratory data and neuroimaging findings of frequent diseases as well as important ones including multiple system atrophies(MSA), drug-induced parkinsonism, vascular pseudo-parkinsonism and manganese intoxication due to parenteral nutrition. MRI is useful to diagnose MSA, vascular pseudo-parkinsonism and manganese intoxication. Benzamide derivatives including sulpiride, tiapride, metoclopramide and cisapride are the main causes of drug-induced parkinsonism in recent years in Japan.

Smythies J. 2000 Fall. Redox aspects of signaling by catecholamines and their metabolites. Antioxid Redox Signal 2(3):575-83.
Abstract: This review covers certain novel aspects of catecholamine signaling in neurons that involve redox systems and synaptic plasticity. The redox hypothesis suggests that one important factor in neurocomputation is the formation of new synapses and the removal of old ones (synaptic plasticity), which is modulated in part by the redox balance at the synapse between reactive oxygen species (ROS) (such as hydrogen peroxide and the nitric oxide radical) and neuroprotective antioxidants (such as ascorbate, glutathione, and catecholamines). Catecholamines, in particular dopamine, which signals positive reinforcement, may play a key role in this

activity. Dopamine has powerful antioxidant properties by several separate mechanisms-direct ROS scavenging, activation of the synthesis of antioxidant proteins, and possibly via dismuting complexes with iron inside endosomes or in catecholaminergic synaptic vesicles. This may contribute to synaptic growth and reinforcement-directed learning. On the other hand, catecholamines are easily oxidized to toxic quinones on the neuromelanin pathway. This might contribute under certain circumstances to synaptic deletion. Evidence is presented that abnormalities in this system may contribute to the pathogenesis of Parkinson's disease and schizophrenia.

Kannan K, Jain SK. 2000 Sep. Oxidative stress and apoptosis. Pathophysiology 7 (3):153-163.
Abstract: Apoptosis or programmed cell death, is essential for the normal functioning and survival of most multi-cellular organisms. The morphological and biochemical characteristics of apoptosis, however, are highly conserved during the evolution. It is currently believed that apoptosis can be divided into at least three functionally distinct phases, i.e. induction, effector and execution phase. Recent studies have demonstrated that reactive oxygen species (ROS) and the resulting oxidative stress play a pivotal role in apoptosis. Antioxidants and thiol reductants, such as N-acetylcysteine, and overexpression of manganese superoxide (MnSOD) can block or delay apoptosis. Bcl-2, an endogenously produced protein, has been shown to prevent cells from dying of apoptosis apparently by an antioxidative mechanism. Taken together ROS, and the resulting cellular redox change, can be part of signal transduction pathway during apoptosis. It is now established that mitochondria play a prominent role in apoptosis. During mitochondrial dysfunction, several essential players of apoptosis, including pro-caspases, cytochrome C, apoptosis-inducing factor (AIF), and apoptotic protease-activating factor-1 (APAF-1) are released into the cytosol. The multimeric complex formation of cytochrome C, APAF-1 and caspase 9 activates downstream caspases leading to apoptotic cell death. All the three functional phases of apoptosis are under the influence of regulatory controls. Thus, increasing evidences provide support that oxidative stress and apoptosis are closely linked physiological phenomena and are implicated in pathophysiology of some of the chronic diseases including AIDS, autoimmunity, cancer, diabetes mellitus, Alzheimer's and Parkinson's and ischemia of heart and brain.

Youdim MB. 2000 Jul-Aug. Nutrient deprivation and brain function: iron. Nutrition 16(7-8):504-8.

Jimenez Del Rio M, Velez-Pardo C. 2000 Jul. 17 beta-estradiol protects lymphocytes against dopamine and iron-induced apoptosis by a genomic-independent mechanism. Implication in Parkinson's disease. Gen Pharmacol 35(1):1-9.
Abstract: Dopamine (DA) in combination with iron (Fe(2+)) has been demonstrated to induce apoptosis in neuronal-like PC12 cells by an oxidative stress mechanism. To get a better insight of cell death and protective mechanisms in DA/Fe(2+)-induced toxicity, we investigated the effects of DA/Fe(2+) and the antioxidant action of 17 beta-estradiol (E2) in peripheral blood lymphocytes (PBL). We found that DA/Fe(2+)-induces apoptosis in PBL via a hydrogen peroxide (H(2)O(2))-mediated oxidative mechanism, which in turn triggers a cascade of molecular events requiring RNA and de novo protein synthesis. We have also demonstrated that E2 prevents significantly DA/Fe(2+)-induced apoptosis in PBL by directly inhibiting the intracellular accumulation of peroxides generated by DA/Fe(2+)-reaction. This protective activity is independent of the presence or activation of the estrogen receptors (ERs). These data further support and validate our previous hypothesis that DA/Fe(2+)/H(2)O(2) could be a general mediator of oxidative stress through a common cell death mechanism in both neuronal and nonneuronal cells. These findings may be particularly relevant to the potential approaches to rescue and prolong the survival of neurons by estrogens in patients with Parkinson's disease (PD).

Hanley KW, Lenhart SW. 2000 Jul. Manganese dioxide exposures and respirator

performance at an alkaline battery plant. Appl Occup Environ Hyg 15(7): 542-9.
Abstract: Two industrial hygiene studies were conducted at an alkaline battery plant to evaluate worker exposures to manganese dioxide particulate and the effectiveness of filtering facepiece respirators. The work areas studied included the plant's powder-processing tower and press rooms where manganese was blended, compacted with graphite, and inserted into battery cans. Full-shift personal breathing zone monitoring was conducted to estimate manganese dust exposures of press operators, mechanics, and material handlers. In-facepiece and personal breathing zone air sampling pairs were also collected using a program protection factor protocol to estimate the protection provided by the respirators. Particle size evaluations were made using nylon cyclones and Marple personal multi-stage impactors. All samples were analyzed for manganese by inductively coupled argon plasma, atomic emission spectroscopy via NIOSH analytical method 7300 utilizing a modified acid digestion procedure. Fifty-four, full-shift, time-weighted average (TWA) exposures to total manganese ranged from 0.1 to 5.4 milligrams per cubic meter (mg/m3); worker exposures were substantially lower during a follow-up study due to engineering control improvements. Concurrent area sample comparisons of total and respirable manganese revealed that the respirable particulate mass fractions ranged from 6 to 32 percent, and mass median aerodynamic diameters determined from personal breathing zone air samples were mostly greater than 10 micrometers. Fifteen respirator performance evaluations were conducted using Moldex 2200 respirators fitted with 25 millimeter cassettes and light weight sampling probes. Protection factors ranged from 5 to 220, with a geometric mean and standard deviation of 31 and 2.97, respectively. The 5th percentile protection factor estimate was 5, as calculated from the protection factor distribution for this sample set. In 1995, the American Conference of Governmental Industrial Hygienists (ACGIH) lowered the elemental and inorganic manganese dust Threshold Limit Value (TLV) from 5 mg/m3 to 0.2 mg/m3 to address adverse pulmonary and central nervous system effects and male infertility. Although most personal breathing zone concentrations were above 0.2 mg/m3, none of the in-facepiece concentrations exceeded this concentration. Parkinson's-like symptoms have been reported in the literature for high manganese dust and fume exposures, but the importance of low dust exposures for producing neurological effects is uncertain.

Youdim MB, Yehuda S. 2000 May. The neurochemical basis of cognitive deficits induced by brain iron deficiency: involvement of dopamine-opiate system. Cell Mol Biol (Noisy-Le-Grand) 46(3):491-500.
Abstract: Iron is an essential element in maintaining normal structure and functions of the central nervous system. Dangerous effects of decreases in the bioavailability of iron in the brain are shown to affect brain biochemistry, neurotransmitters production and function, mainly in the dopamine-opiate systems well as cognitive functions (learning and memory) and a number of physiological variables such motor activity and thermoregulation. Recent research has shown the added complications and deficits that are introduced in the endocrine and the immune system activity. While iron deficiency is not perceived as a life threatening disorder, it is the most prevalent nutritional disorder in the world and a better understanding of the modes and sites of action, can help devise better treatment programs for those who suffer from it.

Kocaturk PA, Akbostanci MC, Tan F, Kavas GO. 2000 Apr. Superoxide dismutase activity and zinc and copper concentrations in Parkinson's disease. Pathophysiology 7(1):63-67.
Abstract: Although several hypotheses are currently being investigated the cause of Parkinson's disease (PD) is still unknown. The aim of this study was to determine red cell copper/zinc-superoxide dismutase (Cu/Zn-SOD) activity and copper and zinc concentrations both in plasma and in red cell in PD. In this preliminary assay, 30 patients with PD the mean age of 64 were studied. Additionally, a second group of older individuals without PD

mean age of 61, were recruited to the study. The patient group was compared with the other group according to their red cell Cu/Zn-SOD activities, and plasma and red cell copper, zinc concentrations. Red cell Cu/Zn-SOD activity was measured spectrophotometrically while plasma and red cell copper, zinc concentrations were determined by atomic absorption spectrophotometer. The results were analysed by 'Student t-test' statistically. The results showed that red cell Cu/Zn-SOD activities and red cell copper and zinc and also plasma copper concentrations of the PD patients increased compared to older individuals without PD. These findings suggested that possibility of oxidative stress in PD was reflected on the blood including the red cell and plasma parameters.

Wu RM, Tai CH, Chen RC. 2000 Mar. Monitoring of the levodopa concentration-response relationship in Parkinson's disease. Kaohsiung J Med Sci 16(3): 117-25.
Abstract: Motor fluctuations and abnormal involuntary movements are common complications encountered in advanced Parkinson's disease (PD) patients with long-term levodopa therapy. Monitoring of plasma levodopa concentrations and clinical effects has been reported to benefit the management of these complications. However, to our knowledge, there is no data available in Taiwan concerning the correlation between the plasma levodopa levels and motor fluctuations. In this study, we developed the laboratory methodology for plasma levodopa determination by using the aluminum extraction procedure and HPLC-ED. Serial blood samples and motor scores were obtained from 7 PD patients, and the correlation between plasma levodopa levels and motor responses were studied individually. In three patients with wearing-off phenomenon, plasma levodopa concentrations are compatible with the clinical "on" and "off" states. In the other four patients with complex fluctuating responses, their levodopa dosages were adjusted by the results of monitoring. Better motor responses were achieved by optimization of the levodopa pharmacokinetics in these patients. Our preliminary data suggest that simultaneous monitoring of plasma levodopa concentrations and clinical effects might be helpful to improve the therapeutic strategy in some of the parkinsonian patients with fluctuating responses to levodopa.

Sulzer D, Zecca L. 2000 Feb. Intraneuronal dopamine-quinone synthesis: a review. Neurotox Res 1(3):181-95.
Abstract: Dopamine-quinone is synthesized by oxidation of the catechol ring of dopamine. If this occurs within the neuronal cytosol, the quinone may react with cytosolic components, particularly with cysteine residues. In contrast, if quinone is produced within neuronal lysosomes it may provide the fundamental building block for neuromelanin. Since the population of neurons that die in Parkinson's disease are those that display obvious intralysosomal neuromelanin and since cytosolic dopamine-dependent oxyradical formation may underlie methamphetamine toxicity and other specific forms of neurodegeneration in dopaminergic neurons, it is important to elucidate the pathways leading to production of dopamine-quinone. Here we review pathways by which intracellular catechols may be oxidized to quinones, either enzymatically or via reduction of ferric iron or other metals. These metabolites can be adduced by cysteine, could underlie aberrant metabolism and ubiquitination pathways, may induce Lewy body formation, and mediate the synthesis of hydroxyl radical and oxyradical species. Finally, we suggest that by accumulating excess cytosolic catecholamine, neuromelanin synthesis may safely sequester quinones that would otherwise be produced in the neuronal cytosol.

Jimenez del Rio M, Velez-Pardo C. 2000 Feb. Molecular mechanism of monoamine toxicity in Parkinson's disease: hypothetical cell death model. Med Hypotheses 54(2):269-74.
Abstract: Although there have been experimental approaches to understanding the etiology of Parkinson's disease, the cause of cell degeneration in this neurological disorder remains a mystery. Herein, a hypothetical model is proposed to explain the mechanism leading neurons to die. The model is based on recent experimental evidence and it

attempts to dissect the actions of dopamine and metal ions as potential triggers for the activation of an ordered cascade of events of the cell death machinery.

Zhang L, Lee T, Wang Y, Soong TW. 2000. Heterologous expression, functional characterization and localization of two isoforms of the monkey iron transporter Nramp2. Biochem J 349:289-297.
Abstract: Natural resistance-associated macrophage protein 2 (Nramp2) has been suggested to be involved in transferrin-independent iron uptake. Two isoforms of the Nramp2 gene generated by alternative splicing of the 3' exons were identified in mouse, rat and human, but it is unclear if they perform distinct functions. To rationalize our previous work, which indicated an increase in iron deposition in a Parkinsonian monkey brain, two monkey Nramp2 isoforms were isolated for a comparative study to assess their relative iron-uptake abilities. tissue distribution and sub-cellular localization. The monkey Nramp2 isoforms, 2a and 2b, exhibit approx, 98% identity at the amino acid level when compared with the human homologues. The Nramp2a transcript contains a canonical iron-responsive element (IRE), whereas that of Nramp2b lacks the IRE motif in the 3' untranslated region, By reverse transcriptase (RT)-PCR, the mRNAs of both isoforms were detected in ail tissues examined. The amino acid differences at the C-terminus neither affected the protein expression levels in HEK-293T and COS-7 cells nor altered the subcellular localization and tissue distribution of the isoforms. Similar levels of iron uptake were detected in the HEK-293T cells transfected with either the Nramp2a or 2b gene, and a reduction of iron from the ferric (Fe3+) to the ferrous (Fe2+) state is necessary before transport can take place. However, this transferrin-independent uptake of iron into the cells is not a Ca2+-dependent process.

Zhang FP, Bi SP, Liu F, Bian NS. 2000. Indirect differential pulse voltammetric determination of aluminum in biological samples in the presence of 3,4-dihydroxyphenylalanine. Analytical Letters 33(2):209-219.
Abstract: Indirect differential pulse voltammetric (DPV) determination of aluminum in the presence of 3,4-dihydroxyphenylalanine (L-dopa) with glass carbon electrode as working electrode has been described. The method relies on the decrease of DPV anodic peak current of L-dopa with the addition of Al-III. The decreasing value of the peak current is linear with the increase of Al-III concentration. Under the optimum experimental conditions (pH 4.8, 6x10(-4) M L-dopa, 0.06M NaAc - HAc (1)buffer solution), the linear ranges are 4.0x10(-7) - 5.2x10(-6) M and 7.2x10(-6) - 4.5x10(-5) M. The relative standard deviation for 8x10(-6) M aluminum is 1.0% (n = 8) and the detection limit is 3.5x10(-7) M. A number of foreign species for interference have been studied. The method has been applied to determine aluminum in drinking water, synthetic renal dialysate and urine samples.

Zhang FP, Bi SP. 2000. Development of the studies on the aluminum intoxication in the pathology of Alzhmer's disease, Parkinson's disease and dialysis diseases and on the drugs unloading aluminum in human body. Chinese Journal of Inorganic Chemistry 16(3):395-403.
Abstract: The development of the studies on the aluminum intoxication in the pathology of Alzhmer's disease, Parkinson's disease, dialysis encephalopathy, osteomalacia and anaemia and on the drugs unloading aluminum in human body in recent decades have been reviewed in this paper. The etiologies and pathophysiology of the interested diseases are discussed. The aluminum unloading drugs involved desferrioxamine, hydroxypyridines, hydroxypyrones, triamino-trideoxyinositols and catechols. Rise, the roles of serum transferrin in transportation and storage of aluminum in the blood are mentioned.

Youdim MBH, Gassen M, Gross A, Mandel S, Grunblatt E. 2000. Iron chelating, antioxidant and cytoprotective properties of dopamine receptor agonist; apomorphine. Journal of Neural Transmission-Supplement (58):83-96.
Abstract: There have been many attempts to discover neuroprotective

drugs for the treatment of Parkinson's disease (PD). Many of these compounds either do not cross the blood brain barrier or are not very effective in the 6-hydroxydopamine or MPTP (N-methyl-4-phenyl-1,2,3,6-terahydropyridine) models of PD. We have examined several compounds including dopamine receptor agonist bromocritine, lisuride, pergolide and R-apomorphine for their neuroprotective action against the above neurotoxins in PC12 and dopamine neuroblastoma cell lines in culture and in vivo. R-apomorphine exhibited relatively potent neuroprotective action in vitro, cell culture and in vivo as a radical scavenger and iron chelator, because of its catechol structure. The recent clinical trials with apomorphine, where parkinsonian subjects can be weaned off L-dopa would suggest that this drug either exerts a neuroprotective action or that continuous sustained stimulation of dopamine receptor may be responsible for its unusual pharmacological activity. Apomorphine has a far more broad neuroprotective activity in the various models as compared with 1-selegiline and may therefore be an ideal drug to study neuroprotection in parkinsonian subjects with the use of PET or SPECT.

Youdim MB, Gassen M, Gross A, Mandel S, Grunblatt E. 2000. Iron chelating, antioxidant and cytoprotective properties of dopamine receptor agonist; apomorphine. J Neural Transm Suppl (58):83-96.
Abstract: There have been many attempts to discover neuroprotective drugs for the treatment of Parkinson's disease (PD). Many of these compounds either do not cross the blood brain barrier or are not very effective in the 6-hydroxydopamine or MPTP (N-methyl-4-phenyl-1,2,3,6-terahydropyridine) models of PD. We have examined several compounds including dopamine receptor agonist bromocritine, lisuride, pergolide and R-apomorphine for their neuroprotective action against the above neurotoxins in PC12 and dopamine neuroblastoma cell lines in culture and in vivo. R-apomorphine exhibited relatively potent neuroprotective action in vitro, cell culture and in vivo as a radical scavenger and iron chelator, because of its catechol structure. The recent clinical trials with apomorphine, where parkinsonian subjects can be weaned off L-dopa would suggest that this drug either exerts a neuroprotective action or that continuous sustained stimulation of dopamine receptor may be responsible for its unusual pharmacological activity. Apomorphine has a far more broad neuroprotective activity in the various models as compared with 1-selegiline and may therefore be an ideal drug to study neuroprotection in parkinsonian subjects with the use of PET or SPECT.

Youdim MB. 2000. Iron deficiency effects on brain function. Public Health Rev 28 (1-4):83-8.

Witholt R, Gwiazda RH, Smith DR. 2000. The neurobehavioral effects of subchronic manganese exposure in the presence and absence of pre-parkinsonism. Neurotoxicol Teratol 22(6):851-861.
Abstract: Recent studies have implicated chronic elevated exposures to environmental agents, such as metals (e.g., manganese, Mn) and pesticides, as contributors to neurological disease. In particular, there is a concern that sensitive subpopulations such as the aged may be at increased risk for the onset of neurologic disorders because elevated exposures to Mn is associated with increased incidence of parkinsonism. Here, we utilized a rat model of pre-parkinsonism to investigate the effects of Mn exposure on neurotoxicity and the exacerbation of parkinsonism. A pre-parkinsonism state was induced using a unilateral intrastriatal injection of 6-hydroxydopamine (6-OHDA), followed 4 weeks later by Mn exposure (4.8 mg Mn/kg x 3 intraperitoneal injections/week) for 5 weeks. Female Sprague-Dawley rats (n = 44) were divided among the following treatments: (A) control, saline/vehicle; (B) Mn only; (C) 6-OHDA only; and (D) 6-OHDA + Mn. Brain Mn levels were measured by ICP MS. Neurobehavioral function was assessed following Mn exposure using a functional observational battery (FOB) consisting of 10 neurobehavioral tests. Unilateral O-OHDA lesions produced significant ipsilateral vs, contralateral striatal dopamine depletions (60-70%), but no measurable impairment of neurobehavioral function, thereby substantiating this pre-

parkinsonism (i.e., subthreshold) model. In contrast, Mn exposure resulted in significant impairment of neurobehavioral function for eight of the 10 FOE tests. No effects of Mn exposure on striatal dopamine depletion were detected, despite the 3.4-fold increase in brain Mn levels over controls. Notably, Mn exposure in the presence of a pre-parkinsonism state significantly exacerbated the neurobehavioral impairment in the reactivity to handling ($P < .049$) and hopping contralateral rear limb ($P < .033$) FOE tests. While the persistence and Mn dose - response relationship of these neurobehavioral effects were not evaluated here, these results nonetheless suggest that chronic Mn exposure may increase the risk of neurobehavioral impairment in subpopulations that are in a pre-parkinsonism state. (C) 2000 Elsevier Science Inc. All rights reserved.

Wissler JH, Logemann E. 2000. Bioinformatics applied to neurodegenerative diseases suggests physiological prion [PrP], Alzheimer's amyloid precursor (APP), Parkinson's parkin [P] and Huntington's huntingtin [H] proteins being [Cu/Zn]-metalloregulated RNA-binding protein families. Mol Biol Cell 11:132A.

Watson WH, Cai JY, Jones DP. 2000. Diet and apoptosis. Annu Rev Nutr 20:485-505.
Abstract: A range of compounds in or derived from the diet modulates apoptosis in cell cultures in vitro. These observations have important implications concerning the mechanisms whereby dietary components affect health. Proapoptotic compounds could protect against cancer by enhancing elimination of initiated, precancerous cells, and antiapoptotic compounds could promote tumor formation by inhibiting apoptosis in genetically damaged cells. Proapoptotic compounds could also contribute to age-related degenerative diseases by activating cell death in postmitotic cells or shifting the normal balance of mitosis and apoptosis in tissues with regenerative capacity. Many age-related diseases, for example macular degeneration and Parkinson's disease, appear to have oxidative stress as an underlying component that interacts with genetic, dietary, and environmental factors to determine relative risk in an individual. Oxidative stress activates apoptosis, and antioxidants protect against apoptosis in vitro; thus, a central role of dietary antioxidants may be to protect against apoptosis. However, little in vivo data are available to directly link diet with altered apoptosis as an underlying determinant of disease. Moreover, the possible antagonistic effects of different dietary components and the uncertainty about whether proapoptotic compounds that may protect against cancer could contribute to degenerative diseases and vice versa indicate that there is a great need for better in vivo assessment of apoptosis and that caution should be exercised when extrapolating in vitro data on apoptosis to in vivo dietary recommendations.

Wakabayashi K, Fukushima T, Koide R, Horikawa Y, Hasegawa M, Watanabe Y, Noda T, Eguchi I, Morita T, Yoshimoto M, Iwatsubo T, Takahashi H. 2000. Juvenile-onset generalized neuroaxonal dystrophy (Hallervorden-Spatz disease) with diffuse neurofibrillary and Lewy body pathology. Acta Neuropathol (Berl) 99(3):331-336.
Abstract: We describe an unusual case of Hallervorden-Spatz disease (HSD). After presenting with limb rigidospasticity at the age of 9 years, our patient developed progressive dementia, spastic tetraparesis and myoclonic movements, leading to akinetic mutism. He died of pneumonia at the age of 39 years. Autopsy revealed a severely atrophic brain, weighing 510 g. Histologically, there were iron deposits in the globus pallidus and substantia nigra pars reticulata, and numerous axonal spheroids throughout out the brain and spinal cord. Neurofibrillary tangles were abundant in the hippocampus, cerebral neocortex, basal ganglia and brain stem. Neuritic plaques and amyloid deposits were absent. Lewy bodies and Lewy neurites, which were immunolabeled by anti-alpha-synuclein, were found in the brain stem, cerebral cortex and spinal gray matter. Sarkosyl-insoluble tau extracted from the temporal cortex resolved on immunoblots into three major bands of 60, 64 and 68 kDa and a minor band of 72 kDa, as reported for Alzheimer's disease. The present case,

together with a few similar cases reported previously, may represent a particular subset of neuroaxonal dystrophy, i.e., HSD associated with extensive accumulation of both tau and alpha-synuclein.

Vymazal J, Urgosik D, Bulte JWM. 2000. Differentiation between hemosiderin- and ferritin-bound brain iron using nuclear magnetic resonance and magnetic resonance imaging. Cell Mol Biol 46(4):835-842.
Abstract: MRI is an optimal clinical (research) tool to provide information on brain morphology and pathology and to detect metal ions that possess intrinsic magnetic properties. Non-heme iron is abundantly present in the brain in three different forms: "low molecular weight" complexes, iron bound to "medium mollecular weight complexes" metalloproteins such as transferrin, and "high molecular weight" complexes as ferritin and hemosiderin. The total amount and form of iron may differ in health and disease, and MRT. can possibly quantify and monitor such changes. Ferritin-bound iron is the main storage form of iron and is present predominantly in the extrapyramidal nuclei where its amounts normally increase as a function of age. Ferritin is water soluble and shortens both, T-1 and T-2 relaxation, with as result a signal change on the MR images. Hemosiderin, a degradation product of ferritin, is water-insoluble with a stronger T-2 shortening effect than ferritin. The larger cluster size of hemosiderin and its water-insolubility also explain a lack of significant T-1-shortening effect on T-1-weighted images. Using both in vitro specimens and intact brain tissue in vivo we demonstrate here that MRT may be able to distinguish between ferritin- and hemosiderin-bound iron.

Trombley PQ, Horning MS, Blakemore LJ. 2000. Interactions between carnosine and zinc and copper: Implications for neuromodulation and neuroprotection. Biochemistry-Moscow 65(7):807-816.
Abstract: This review examines Interactions in the mammalian central nervous system (CNS) between carnosine and the endogenous transition metals zinc and copper. Although the relationship between these substances may be applicable to other brain regions, the focus is on the olfactory system where these substances may have special significance. Carnosine is not only highly concentrated in the olfactory system, but it is also contained in neurons (in contrast to glia cells in most of the brain) and has many features of a neurotransmitter, Whereas the function of carnosine in the CNS is not well understood, we review evidence that suggests that it may act as both a neuromodulator and a neuroprotective agent. Although zinc and/or copper are found in many neuronal pathways in the brain, the concentrations of zinc and copper in the olfactory bulb (the target of afferent input from sensory neurons in the nose) are among the highest in the CNS, Included in the multitude of physiological roles that zinc and copper play in the CNS is modulation of neuronal excitability, However, zinc and copper also have been implicated in a variety of neurologic conditions including Alzheimer's disease, Parkinson's disease, stroke, and seizures. Here we review the modulatory effects that carnosine can have on zinc and copper's abilities to. influence neuronal excitability and to exert neurotoxic effects in the olfactory system. Other aspects of carnosine in the CNS are reviewed elsewhere in this issue.

Torsdottir G, Kristinsson J, Gudmundsson G, Snaedal J, Johannesson T. 2000. Copper, ceruloplasmin and superoxide dismutase (SOD) in amyotrophic lateral sclerosis. Pharmacology & Toxicology 87(3):126-130.
Abstract: In two previous studies we found copper dyshomeostasis in patients with Alzheimer's disease and in patients with Parkinson's disease. In this study, the levels of copper in plasma, of ceruloplasmin in serum, ceruloplasmin oxidative activity, ceruloplasmin specific oxidative activity (activity related to mass) as well as superoxide dismutase (SOD) activity in erythrocytes have been determined in 14 patients with amyotrophic lateral sclerosis and their healthy age- and gender-mathced controls. Three of the patients had a familial form of the disease or were suspected of having it. The mean values of all parameters were found not to differ significantly between the patients and their controls (Student's t-test; $P>0.05$). By testing the equality of variances (F distribution) we found that the

variances of individual results fur ceruloplasmin specific oxidative activity and SOD activity differed significantly between the patients group and the controls group (P= 0.021 and P=0.003), but the individual results of these two activities were not correlated (P>0.05). We conclude that disturbances in ceruloplasmin specific oxidative activity and SOD activity could contribute to motor neurone death in amyotrophic lateral sclerosis, and since the two enzyme activities are not correlated it is uncertain which one is more closely related to the pathology of the disease.

Thiruchelvam M, Richfield EK, Baggs RB, Tank AW, Cory-Slechta DA. 2000. The nigrostriatal dopaminergic system as a preferential target of repeated exposures to combined paraquat and maneb: Implications for Parkinson's disease. J Neurosci 20(24):9207-9214.
Abstract: Experimental evidence supporting 1,1'-dimethyl-4,4'-bipyridinium [paraquat (PQ)] as a risk factor for Parkinson's disease (PD) is equivocal. Other agricultural chemicals, including dithiocarbamate fungicides such as manganese ethylenebisdithiocarbamate [maneb (MB)], are widely used in the same geographical regions as paraquat and also impact dopamine systems, suggesting that mixtures may be more relevant etiological models. This study therefore proposed that combined PQ and MB exposures would produce greater effects on dopamine (DA) systems than would either compound administered alone. Male C57BL/6 mice were treated twice a week for 6 weeks with intraperitoneal saline, 10 mg/kg paraquat, 30 mg/kg maneb, or their combination (PQ 1 MB). MB, but not PQ, reduced motor activity immediately after treatment, and this effect was potentiated by combined PQ 1 MB treatment. As treatments progressed, only the combined PQ 1 MB group evidenced a failure of motor activity levels to recover within 24 hr. Striatal DA and dihydroxyphenylacetic acid increased 1-3 d and decreased 7 d after injections. Only PQ 1 MB reduced tyrosine hydroxylase (TH) and DA transporter immunoreactivity and did so in dorsal striatum but not nucleus accumbens. Correspondingly, striatal TH protein levels were decreased only by combined PQ 1 MB 5 d after injection. Reactive gliosis occurred only in response to combined PQ 1 MB in dorsal-medial but not ventral striatum. TH immunoreactivity and cell counts were reduced only by PQ 1 MB and in the substantia nigra but not ventral tegmental area. These synergistic effects of combined PQ 1 MB, preferentially expressed in the nigrostriatal DA system, suggest that such mixtures could play a role in the etiology of PD.

Takanashi M, Mochizuki H, Hattori N, Mori H, Mizuno Y. 2000. Iron accumulation in the substantia nigra of autosomal recessive juvenile parkinsonism. Brain Pathol 10(4):782.

Sulzer D, Bogulavsky J, Larsen KE, Behr G, Karatekin E, Kleinman MH, Turro N, Krantz D, Edwards RH, Greene LA, Zecca L. 2000. Neuromelanin biosynthesis is driven by excess cytosolic catecholamines not accumulated by synaptic vesicles. Proc Natl Acad Sci U S A 97(22):11869-11874.
Abstract: Melanin, the pigment in hair, skin, eyes, and feathers, protects external tissue from damage by UV light. In contrast, neuromelanin (NM) is found in deep brain regions, specifically in loci that degenerate in Parkinson's disease. Although this distribution suggests a role for NM in Parkinson's disease neurodegeneration, the biosynthesis and function of NM have eluded characterization because of lack of an experimental system. We induced NM in rat substantia nigra and PC12 cell cultures by exposure to L-dihydroxy-phenylalanine, which is rapidly converted to dopamine (DA) in the cytosol. This pigment was identical to human NM as assessed by paramagnetic resonance and was localized in double membrane autophagic vacuoles identical to NM granules of human substantia nigra. NM synthesis was abolished by adenoviral-mediated overexpression of the synaptic vesicle catecholamine transporter VMAT2, which decreases cytosolic DA by increasing vesicular accumulation of neurotransmitter. The NM is in a stable complex with ferric iron, and NM synthesis was inhibited by the iron chelator desferrioxamine, indicating that cytosolic DA and dihydroxyphenylalanine are oxidized by iron-mediated

catalysis to membrane-impermeant quinones and semiquinones. NM synthesis thus results from excess cytosolic catecholamines not accumulated into synaptic vesicles. The permanent accumulation of excess catechols, quinones, and catechol adducts into a membrane-impermeant substance trapped in organelles may provide an antioxidant mechanism for catecholamine neurons. However, NM in organelles associated with secretory pathways may interfere with signaling, as it delays stimulated neurite outgrowth in PC12 cells.

Strelau J, Sullivan A, Bottner M, Lingor P, Falkenstein E, Suter-Crazzolara C, Galter D, Jaszai J, Krieglstein K, Unsicker K . 2000. Growth/differentiation factor-15/macrophage inhibitory cytokine-1 is a novel trophic factor for midbrain dopaminergic neurons in vivo. J Neurosci 20(23):8597-8603.
Abstract: Transforming growth factor-betas (TGF-betas) constitute an expanding family of multifunctional cytokines with prominent roles in development, cell proliferation, differentiation, and repair. We have cloned, expressed, and raised antibodies against a distant member of the TGF-betas, growth/differentiation factor-15 (GDF-15). GDF-15 is identical to macrophage inhibitory cytokine-1 (MIC-1). GDF-15/MIC-1 mRNA and protein are widely distributed in the developing and adult CNS and peripheral nervous systems, including choroid plexus and CSF. GDF-15/MIC-1 is a potent survival promoting and protective factor for cultured and iron-intoxicated dopaminergic (DAergic) neurons cultured from the embryonic rat midbrain floor. The trophic effect of GDF-15/MIC-1 was not accompanied by an increase in cell proliferation and astroglial maturation, suggesting that GDF-15/MIC-1 probably acts directly on neurons. GDF-15/MIC-1 also protects 6-hydroxydopamine (6-OHDA)-lesioned nigrostriatal DAergic neurons in vivo. Unilateral injections of GDF-15/MIC-1 into the medial forebrain bundle just above the substantia nigra (SN) and into the left ventricle (20 mug each) immediately before a 6-OHDA injection (8 mug) prevented 6-OHDA-induced rotational behavior and significantly reduced losses of DAergic neurons in the SN. This protection was evident for at least 1 month. Administration of 5 mug of GDF-15/MIC-1 in the same paradigm also provided significant neuroprotection. GDF-15/MIC-1 also promoted the serotonergic phenotype of cultured raphe neurons but did not support survival of rat motoneurons. Thus, GDF-15/MIC-1 is a novel neurotrophic factor with prominent effects on DAergic and serotonergic neurons. GDF-15/MIC-1 may therefore have a potential for the treatment of Parkinson's disease and disorders of the serotonergic system.

Stokes AH, Lewis DY, Lash LH, Jerome WG, Grant KW, Aschner M, Vrana KE. 2000. Dopamine toxicity in neuroblastoma cells: role of glutathione depletion by L-BSO and apoptosis. Brain Res 858(1):1-8.
Abstract: Dopamine (DA), while an essential neurotransmitter, is also a known neurotoxin that potentially plays an etiologic role in several neurodegenerative diseases. DA metabolism and oxidation readily produce reactive oxygen species (ROS) and DA can also be oxidized to a reactive quinone via spontaneous, enzyme-catalyzed or metal-enhanced reactions. A number of these reactions are cytotoxic, yet the precise mechanisms by which DA leads to cell death remain unknown. In this study, the neuroblastoma cell line, SK-N-SH, was utilized to examine DA toxicity under varying oxidant states. Cells pretreated with the glutathione (GSH)-depleting compound, L-buthionine sulfoximine (L-BSO), exhibited enhanced sensitivity to DA compared to controls (non-GSH-depleted cells). Furthermore, in cells pretreated with L-BSO, the addition of ascorbate (250 mu M) afforded significant protection against DA-induced toxicity, while pyruvate (500 mu M) had no protective effect. To further characterize the possibility that DA is associated with oxidative stress, additional studies were carried out with manganese (30 mu M) as a pro-oxidant. Manganese and DA (200 mu M); although not cytotoxic when individually administered to SK-N-SH cells, had a synergistic action on cytotoxicity. Finally, morphological and molecular markers of programmed cell death (apoptosis) were observed in cells treated with DA and L-BSO. These markers included membrane blebbing and internucleosomal DNA fragmentation. These results suggest that DA toxicity is tightly linked to

intracellular oxidant/antioxidant levels, and that environmental factors, such as excessive Mn exposure, may modulate cellular sensitivity to DA. (C) 2000 Published by Elsevier Science B.V. All rights reserved.

Spahr L, Lazeyras F, Dupasquier R, Delavelle J, Burkhard P, Hadengue A, Hochstrasser D, Mentha G, Terrier F, Vingerhoets F. 2000. Evolution of parkinsonian signs, blood manganese, brain magnetic resonance imaging and spectroscopic alterations after liver transplantation in patients with cirrhosis. Hepatology 32(4):521A.

Spadoni F, Stefani A, Morello M, Lavaroni F, Giacomini P, Sancesario G. 2000. Selective vulnerability of pallidal neurons in the early phases of manganese intoxication. Exp Brain Res 135(4):544-551.
Abstract: Prolonged exposure to manganese in mammals may cause an extrapyramidal disorder characterized by dystonia and rigidity. Gliosis in the pallidal segments underlies the well-established phase of the intoxication. The early phase of the intoxication may be characterized by psychic, nonmotor signs, and its morphological and electrophysiological correlates are less defined. In a rat model of manganese intoxication (20 mg/ml in drinking water for 3 months), neither neuronal loss nor gliosis was detected in globus pallidus (GP). However, a striking vulnerability of manganese-treated GP neurons emerged. The majority of GP neurons isolated from manganese-treated rats died following brief incubation in standard dissociation media. In addition, patch-clamp recordings in the whole-cell configuration were not tolerated by surviving GP neurons. Neither coeval but untreated GP neurons nor striatal ones manifested analogous susceptibility. Using the perforated-patch mode of recording we attempted at identifying the functional hallmarks of GP vulnerability: in particular, voltage-gated calcium currents and glutamate-induced currents were examined. Manganese-treated GP neurons exhibited calcium currents similar to control cells aside from a slight reduction in the dihydropyridine-sensitive current facilitation. Strikingly, manganese-treated GP cells - but not striatal ones - manifested peculiar responses to glutamate, since repeated applications of the excitatory amino acid, at concentrations which commonly promote desensitizing responses, produced instead an irreversible cell damage. Possible mechanisms are discussed.

Sole-Garcia MD. 2000. Epidemiological aspects of occupational neurology. Investigation methodology. Rev Neurol 31(9):861-864.
Abstract: Introduction. In the working population, unsatisfactory working conditions (in the broadest sense) may cause effects whose gravity depends mainly on the characteristics of exposure and the general condition of the individual. Development. These effects may, in practice, be classified as occupational accidents and professional illness (both legal concepts), illnesses related to work, diminished quality of life and effects on offspring Agents as varied as head injuries and metals are known to cause from loss of memory or behavior disorders to chronic toxic encephalopathy or Parkinsonism. There is little data on exposure to neurotoxic agents or disorders of the central nervous system caused by work and what there is not reliable. Therefore it is necessary to facilitate and encourage collaboration between neurologists and occupational medicine specialists so as to determine the importance/extent of the problem, undertake investigation as to aetiology and develop methods for early detection of these disorders.

Sogawa CA, Miyazaki I, Sogawa N, Asanuma M, Ogawa N, Furuta H. 2000. Antioxidants protect against dopamine-induced metallothionein-III (GIF) mRNA expression in mouse glial cell line (VR-2g). Brain Res 853(2): 310-316.
Abstract: Metallothionein (MT)-III, originally discovered as a growth inhibitory factor (GIF), is a brain specific isomer of MTs and is markedly reduced in the brain of patients with Alzheimer's disease (AD) or other neurodegenerative diseases. We analyzed the level and regulation of mRNA expression of MT-III in immortalized fetal mouse brain glial cells (VR-2g) by reverse transcriptase-polymerase chain reaction (RT-PCR). We

have recently reported that dopamine (DA) increases the expression of MT-III mRNA in vitro. In this study, we investigated the mechanism of such increase by examining the effects of DA agonists (SKF38393 or bromocriptine) and DA antagonists (SCH23390 or sulpiride) on the expression of MT-III mRNA. MT-III mRNA did not change by either agonist and DA-increased MT-III mRNA was not inhibited by either antagonist. These results suggested that the induction of MT-III mRNA by DA was not mediated by stimulation of DA receptors. On the other hand, DA-induced MT-III mRNA expression was strongly inhibited by the addition of antioxidants (glutathione, vitamin E or ascorbic acid), indicating thar DA-enhanced MT-III mRNA was mediated by reactive oxygen species. Our results suggest that oxidative stress may be one of the principle factors that modulate MT-III mRNA expression. (C) 2000 Elsevier Science B.V. All rights reserved.

Shoham S, Youdim MBH. 2000. Iron involvement in neural damage and microgliosis in models of neurodegenerative diseases. Cell Mol Biol 46(4): 743-760.
Abstract: In several neurodegenerative diseases, iron accumulates at sites of brain pathology. Since post-mortem examination cannot distinguish whether iron accumulation caused the damage or resulted from damage, it is necessary to manipulate iron in animal and tissue culture models to assess its causal role(s). However, only in models of Parkinson's disease and of global ischemia, iron deprivation (ID) or iron-chelators have been used to protect from damage. In these studies, documentation of microgliosis was not performed even though several lines of evidence converge to suggest that activation of microglia is an important source of oxidative stress. In the kainate model of epilepsy, we found that ID protected the olfactory cortex, thalamus and hippocampus and attenuated microgliosis, whereas iron supplementation to TD rats increased damage and microgliosis in the above regions. In the hilus of the hippocampal dentate gyros, even though no cell loss was observed, ID attenuated microgliosis and iron-supplementation increased it. Thus there is a tight relationship between iron and microgliosis. In addition, iron+zine supplementation dramatically increased damage to hippocampal CAl whereas zinc supplementation alone had no effect. This study demonstrates an anatomically unique interaction of iron and zinc, which may lead to new insights to neurodegneration in epilepsy.

Shils JL, Patterson T, Stecker MM. 2000. Electrical properties of metal microelectrodes. American Journal of Electroneurodiagnostic Technology 40(2):143-156.
Abstract: As metal microelectrode use increases, especially in intraoperative procedures, there is a need for greater understanding of the intrinsic signal and noise properties of these electrodes. In this communication we explore some intrinsic noise levels in the electrode-amplifier system and its frequency response function. Noise levels were studied in the presence of a grounded Faraday cage, an ungrounded Faraday cage, and an open electrode-amplifier system, In the 100-2000 Hz frequency range, total noise power in the grounded Faraday cage was 44 mu V, compared to 168 mu V for the ungrounded Faraday cage and 525 mu V without any shielding. The Faraday cage appeared to reduce most of the low frequency noise and grounding reduced mostly line noise. This noise was still in the region of 1000 times greater than the noise in other electrode-amplifier systems. The frequency response of the metal microelectrode system is fairly consistent above 1000 Hz but decreases sharply below 1000 Hz, showing approximately -15 dB between 100 and 1000 Hz. Even though these measured results are fairly mediocre compared to other electrode-amplifier systems, in practice they are still acceptable because single-cell recording amplitudes (intraoperatively, with human subjects) are on the order of 100 to 10,000 times greater than other recordings such as evoked potentials and EEG.

Serra PA, Esposito G, Enrico P, Mura MA, Migheli R, Delogu MR, Miele M, Desole MS, Grella G, Miele E. 2000. Manganese increases L-DOPA auto-oxidation

in the striatum of the freely moving rat: potential implications to L-DOPA long-term therapy of Parkinson's disease. Br J Pharmacol 130(4):937-945.
Abstract: 1 We have previously shown that manganese enhances L-dihydroxyphenylanine (L-DOPA) toxicity to PC12 cells in vitro. The supposed mechanism of manganese enhancing effect [an increase in L-DOPA and dopamine (DA) auto-oxidation] was studied using microdialysis in the striatum of freely moving rats. 2 Systemic L-DOPA [25 mg kg(-l) intraperitoneally (i.p.) twice in a 12 h interval] significantly increased baseline dialysate concentrations of L-DOPA, dihydroxyphenylacetic acid (DOPAC), homovanillic acid (HVA) and uric acid, compared to controls. Conversely, DA and ascorbic acid concentrations were significantly decreased. 3 A L-DOPA oxidation product, presumptively identified as L-DOPA semiquinone, was detected in the dialysate. The L-DOPA semiquinone was detected also following intrastriatal infusion of L-DOPA. 4 In rats given L-DOPA i.p., intrastriatal infusion of N-acetylcysteine (NAC) significantly increased DA and L-DOPA dialysate concentrations and lowered those of L-DOPA semiquinone; in addition, NAC decreased DOPAC + HVA and uric acid dialysate concentrations. 5 In rats given L-DOPA either systemically or intrastriatally, intrastriatal infusion of manganese decreased L-DOPA dialysate concentrations and greatly increased those of L-DOPA semiquinone. These changes were inhibited by NAC infusion. 6 These findings demonstrate that auto-oxidation of exogenous L-DOPA occurs in vivo in the rat striatum. The consequent reactive oxygen species generation may account for the decrease in dialysate DA and ascorbic acid concentrations and increase in enzymatic oxidation of xanthine and DA. L-DOPA auto-oxidation is inhibited by NAC and enhanced by manganese. These results may be of relevance to the L-DOPA long-term therapy of Parkinson's disease.

Schulmann-Choron N, Chevallier P, De Ceaurriz J, Souleau C. 2000. Determination of selenium in rat brain by synchrotron radiation X-ray fluorescence. Analusis 28(4):316-323.
Abstract: Selenium, an ubiquitous essential trace-element is known to be particularly difficult to measure especially in brain. First, it is a non metal, next, at very low concentration (below ppm). at last, the brain matter very rich in lipids. make the digestion specially uneasy. Using synchrotron radiation induced X-ray fluorescence analysis (SXRF), selenium in rat brain was measured equal to C-Se = 124 +/- 5.4 ppb with a MDL (minimum detection limit) of 20 ppb. The obtained values should be used as a first step to study human brain on extremely small and specific locations. With the development of aging pathologies (such as Alzheimers or Parkinson's disease...), it matters to get informations about selenium known as an anti-aging element. The obtained values from rat, near of man's. may also highlight it as a potential animal model system for studying selenium in human brain.

Schipper HM. 2000. Heme oxygenase-1: role in brain aging and neurodegeneration. Exp Gerontol 35(6-7. Sp. Iss. Si):821-830.
Abstract: The mechanisms responsible for excessive iron deposition and mitochondrial insufficiency in the aging and degenerating nervous system remain poorly understood. Heme oxygenase-1 (HO-1) is a 32 kDa stress protein that degrades heme to biliverdin, free iron and carbon monoxide. Our laboratory has shown that cysteamine, dopamine, beta -amyloid, IL-1 beta and TNF-alpha up-regulate HO-1 followed by mitochondrial sequestration of non-transferrin-derived Fe-55 in cultured rat astroglia. In these cells and in rat astroglia transfected with the human HO-1 gene, mitochondrial iron trapping is abrogated by the HO-1 inhibitors, tin-mesoporphyrin and dexamethasone. We determined that HO-1 immunoreactivity is enhanced greatly in neurons and astrocytes of the hippocampus and cerebral cortex of Alzheimer subjects and co-localizes to senile plaques and neurofibrillary tangles (NFT). HO-1 staining is also augmented in astrocytes and decorates neuronal Lewy bodies in the Parkinson nigra. Collectively, our findings suggest that HO-1 over-expression contributes to the pathological iron deposition and mitochondrial damage documented in these aging-related neurodegenerative disorders.

We recently observed that, paradoxically, HO-1 mRNA levels are markedly suppressed in peripheral lymphocytes of patients with early sporadic Alzheimer disease and may thus provide a useful biological marker of this condition. (C) 2000 Elsevier Science Inc. All rights reserved.

Sayre LM, Perry G, Atwood CS, Smith MA. 2000. The role of metals in neurodegenerative diseases. Cell Mol Biol 46(4):731-741.
Abstract: There is increasing evidence in a number of neurodegenerative diseases that transition metal-mediated abnormalities play a crucial role in disease pathogenesis. In this treatise, we review the role of metal homeostasis as it pertains to alterations in brain function in neurodegenerative diseases. In fact, while there is documented evidence for alterations in transition metal homeostasis, redox-activity and localization, it is also important to realize that alterations in specific copper- and iron-containing metalloenzymes also appear to play a crucial role in the neurodegenerative process.

Santiago M, Matarredona ER, Granero L, Cano J, Machado A. 2000. Neurotoxic relationship between dopamine and iron in the striatal dopaminergic nerve terminals. Brain Res 858(1):26-32.
Abstract: The neurotoxic effect of dopamine (DA) and iron(III) on DAergic terminals in striatum has been studied by intracerebral microdialysis technique. Twenty-four hours after surgery (day 1), DA and/or iron(III) with and without DA reuptake inhibitor, nomifensine, were perfused for 1 h. Forty-eight hours after surgery (day 2), MPP+ 1 mM was perfused for 15 min and the output of DA was measured, its amount being directly proportional to the remaining striatal DAergic terminals, supported by tyrosine hydroxylase immunohistochemistry technique. Perfusion of exogenous DA, as well as iron(III) 10 and 100 mu M, did not produce any neurotoxic effect. However, perfusion of iron(III) (333 and 1000 mu M) produced a concentration-dependent toxic effect. Co-perfusion of iron(III) at non-toxic concentration (100 mu M) with DA (15 mu M) produced a toxic effect. Elevation of the endogenous extracellular levels of DA by inhibiting its uptake with nomifensine increased the neurotoxic effect of iron(III) in a dose-dependent manner. The use of tetrodotoxin after elevation of DA with nomifensine partially prevented the neurotoxic effect of its co-perfusion with iron(III) (100 mu M). These results suggest that DAergic system could be synergistically damaged by DA and iron(III). Thus, alterations in the clearance of DA from extracellular space along with an increase of iron may have significant consequences for DAergic system toxicity. (C) 2000 Elsevier Science B.V. All rights reserved.

Samir AM. 2000. The response of benthic foraminifera and ostracods to various pollution sources: A study from two lagoons in Egypt. Journal of Foraminiferal Research 30(2):83-98.
Abstract: A study of foraminiferal assemblages was carried out at two Egyptian Nile Delta lagoons. Analysis of surficial sediment samples from Manzalah Lagoon shows enrichment in heavy metals (Pb, Zn, Cu, Cr and Cd). The environment has become so lethal to foraminifera that no species can currently survive. Among ostracods, only one species (Cyprideis torosa) was found living and able to invade the polluted lagoon region. Samples from Edku Lagoon, which receives only agricultural drainage water, show heavy metal concentrations close to natural baseline levels, and yield living foraminifera, The frequent occurrence of deformed specimens in Manzalah Lagoon, comparable to Edku Lagoon, reveals that: (1) benthic foraminifera are more sensitive to industrial wastes containing heavy metals; (2) agricultural wastes do not significantly harm benthic foraminifera; (3) Ammonia beccarii forma parkinsoniana is less resistant to pollution than forma tepida; (4) morphological abnormalities of the foraminiferal tests depend upon the nature of the pollutant; and (5) benthic foraminifera are less tolerant to pollution than ostracods and molluscs.

Rybicki B, Gorell J, Johnson C, Peterson E, Fisher J, Kirkey K. 2000. Issues related to studying occupational metal exposures and Parkinson's disease.

Epidemiology 11(4):S150.

Roth JA, Feng L, Walowitz J, Browne RW. 2000. Manganese-induced rat pheochromocytoma (PC12) cell death is independent of caspase activation. J Neurosci Res 61(2):162-171.
Abstract: Manganese (Mn) is an essential mineral that at high concentrations can produce an irreversible syndrome resembling Parkinson's disease. To examine the mechanism by which Mn elicits its toxic response, we have selected the rat pheochromocytoma cells (PC12) as our model system because it possesses much of the biochemical machinery associated with dopaminergic neurons. Win-induced PC12 cell death is both time and concentration dependent with approximately 50% cell survival at 48 hr in the presence of 0.3 mM Mn. To determine whether oxidative stress contributed to cytotoxicity induced by Mn, lipid peroxidation was assessed in Mn-treated in PC12 cells. The highly sensitive HPLC assay that measures the lipid peroxide product, 9-HODE, was used and results of these experiments demonstrate there was no increase in the lipid peroxidation in cells exposed to 0.3 mM Mn for 24 hr. Mn was found to stimulate the activation of the apoptotic marker proteins, p38 and caspase-3 within the first 24 hr of treatment. The selective inhibitor of caspase-3, DEVD-CHO, and the nonselective caspase inhibitor, Z-VAD-FMK, however, fail to prevent Mn-induced PC12 cell death. Studies were performed to determine the role of mitochondria in initiating or supporting Mn cytotoxicity, because Mn has been reported to cause changes in membrane permeability. Mn caused a decrease in ATP levels in PC12 cells in both a time and concentration dependent manner. We hypothesize that both apoptosis and necrosis contribute to PC12 cell death although the necrotic events prevail even when the apoptotic signaling is inhibited. (C) 2000 Wiley-Liss, Inc.

Rojas P, Rojas-Castaneda J, Vigueras RM, Habeebu SSM, Rojas C, Rios C, Ebadi M. 2000. MPTP decreases MT-I mRNA in mouse striatum . Neurochem Res 25(4):503-509.
Abstract: 1-Methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) is a drug that induces parkinsonism in humans and non-human primates. Free radicals are thought to be involved in its mechanism of action. Recently, metallothionein has been proposed to play a role as a scavenger of free radicals. In the present work, we studied the effect of MPTP neurotoxicity on brain metallothionein-I (MT-I) mRNA expression. Male C-57 black mice were treated with MPTP (30 mg/kg, i.p., daily) for 3 or 5 days. All animals were killed by cervical dislocation 7 days after the last MPTP dose. The brains were removed quickly and immediately frozen, and quantitative in situ hybridization was performed using MT-I cDNA probe. MT-I mRNA content in striatum, a region which is known to be highly predisposed and sensitive to MPTP-induced oxidative stress, decreased by 30% (3 days) and 39% (5 days) respectively, after the last MPTP administration. These results suggest that MT-I gene expression is decreased in MPTP neurotoxicity. it is suggested that the reduction of MT, an anti-oxidant and a free radical scavenger, in the striatum by MPTP enables the neurotoxin to exert maximal oxidative damage to the striatum.

Rojas P, Hidalgo J, Ebadi M, Rios C. 2000. Changes of metallothionein I+II proteins in the brain after 1-methyl-4-phenylpyridinium administration in mice. Progress in Neuro-Psychopharmacology & Biological Psychiatry 24 (1):143-154.
Abstract: 1. 1-methyl-4-phenylpyridinium (MPP+) is a drug that induces a Parkinson's-like syndrome in several species. Oxidative stress resulting from either excess generation or reduced scavenging of free radicals has been proposed to play a role in its neurotoxicity. 2. It has been suggested that metallothionein (MT) protects against oxidative damage of the central nervous system produced by overproduction of free radicals. 3. This study examined the effect of MPP+ on metallothionein I+II protein content in different brain regions. 4. NIH mice were injected with MPP+ (4.5, 9.0 or 18 mu g/ 3 mu l) into their right lateral ventricle. 5. Corpus striatum, cerebellum, midbrain, frontal cortex and hippocampus were dissected out

and their metallothionein concentrations were analyzed by radioimmunoassay. 6. MPP+ reduced the concentration of MT I+II proteins (38%) only in the striatum. 7. The results suggest that changes in MT I+II content may be associated with MPP+ neurotoxicity.

Ranasinghe JGS, Liu MC, Sakakibara Y, Suiko M. 2000. Manganese administration induces the increased production of dopamine sulfate and depletion of dopamine in Sprague-Dawley rats. J Biochem (Tokyo) 128(3):477-480.
Abstract: Sprague-Dawley rats were used as an experimental model for investigating the effects of manganese poisoning on the serum levels of unsulfated and sulfated forms of dopamine and its biosynthetic precursors, L-Dopa and L-p-tyrosine. Groups of rats were treated daily with Mn2+ (20 mg or 40 mg; in the form of MnSO4) or Na+ (20 mg; in the form of Na2SO4). High performance liquid chromatography (HPLC) analysis of the serum samples taken after a 50-day experimental period revealed that the serum level of dopamine sulfate increased by more than 10 times compared with untreated control rats or rats treated with sodium sulfate. In contrast, there was a dramatic decrease (by as much as 4.8 times) in the serum level of unsulfated dopamine in manganese-treated rats. The serum levels of L-Dopa sulfate and L-p-tyrosine sulfate were also markedly elevated, although not as much as those of dopamine sulfate. Meanwhile, the serum levels of unsulfated L-Dopa and L-p-tyrosine showed no dramatic changes. Atomic absorption spectrophotometric analysis revealed in general an accumulation of manganese in the four organ samples taken from manganese-treated rats. Compared with liver, heart, and kidney, the highest degree of manganese accumulation in manganese-treated rats appeared to be in brain. These results together suggested a role for manganese in stimulating the dopamine-sulfating sulfotransferases in brain, thereby leading to the depletion of dopamine in vivo.

Raha S, Mceachern GE, Myint AT, Robinson BH. 2000. Superoxides from mitochondrial complex III: The role of manganese superoxide dismutase. Free Radic Biol Med 29(2):170-180.
Abstract: In this report we show that ubiquinone cytochrome c reductase (complex III) from isolated rat heart mitochondria when inhibited with antimycin A, produces a large amount of superoxide as measured by the chemiluminescent probe coelenterazine. When mitochondria are inhibited with myxothiazol or stigmatellin, there is no detectable formation of superoxide. The antimycin A-sensitive free radical production can be dramatically reduced using either myxothiazol or stigmatellin. This suggests that the antimycin A-sensitive generation of superoxides originates primarily from the Q(o) semiubiquinone. When manganese superoxide dismutase depleted submitochondrial particles (SMP) were inhibited with myxothiazol or stigmatellin, a large superoxide signal was observed. These two inhibitors likely increase the concentration of the Q(i) semiquinone at the N center. The antimycin A-sensitive signal can, in the case of both the mitochondria and the SMP, be dissipated by the addition of copper zinc superoxide dismutase, suggesting that the measured coelenterazine signal was a result of superoxide production. Taken together, this data suggests that free radicals generated from the Q(i) species are more effectively eliminated by MnSOD in intact mitochondria. (C) 2000 Elsevier Science Inc.

Qian ZM, Liao QK, To Y, Ke Y, Tsoi YK, Wang GF, Ho KP. 2000. Transferrin-bound and transferrin-free iron uptake by cultured rat astrocytes. Cell Mol Biol 46 (3):541-548.
Abstract: Previously we had demonstrated the presence of transferrin receptor (TfR) on the plasma membrane of cultured I at cortical astrocytes. In this study, we investigated the roles of TfR in transferrin-bound iron (Tf-Fe) as well as transferrin-free iron (Fe II) uptake by the cells. The cultured rat astrocytes were incubated with 1 mu M of double-labelled transferrin (I-125-Tf-Fe-59) in serum-free DMEM/F12 medium or Fe-59 II in isotonic sucrose solution at 37 degrees C or 4 degrees C for varying times. The cellular Tf-Fe, Tf and Fe 1T uptake was analyzed by measuring the intracellular radioactivity with gamma counter The result

showed that Tf-Fe uptake kept increasing in a linear manner at least in the first 30-min. In contrast to Tf-Fe uptake, the internalization of Tf into the cells was rapid initially but then slowed to a plateau level after 10 min, of incubation. The addition of either NH4Cl or CH3NH2, the blockers of Tf-Fe uptake via inhibiting iron release from Tf within endosomes, decreased the cellular Tf-Fe uptake but had no significant effect on Tf uptake. Pre-treated cells with trypsin inhibited significantly the cellular uptake of Tf-Fe as well as Tf. These findings suggested that Tf-Fe transport across the membrane of astrocytes is mediated by Tf-TfR endocytosis. The results of transferrin-free iron uptake indicated that the cultured rat cortical astrocytes had the capacity to acquire Fe II. The highest uptake of Fe II occurred at pH 6.5. The Fe II uptake was time and temperature dependent, iron concentration saturable, inhibited by several divalent metal ions, such as Co2+, Zn2+, Mn2+ and Ni2+ and not significantly affected by phenylarsine oxide treatment. These characteristics of Fe II uptake by the cultured astrocytes suggested that Fe II uptake is not mediated by TW and implied that a carrier-mediated iron transport system might be present on the membrane of the cultured cells.

Preat A, Mamet B, De Ridder C, Boulvain F, Gillan D. 2000. Iron bacterial and fungal mats, Bajocian stratotype (Mid-Jurassic, northern Normandy, France). Sedimentary Geology 137(3-4):107-126.
Abstract: The Oolithe ferrugineuse de Bayeux Formation is located at the historical Bajocian stratotype of Sainte-Honorine-des-Pertes, north of Bayeux, Normandy. The condensed formation ranges from the base of the Humphriesianum Zone to the Parkinsoni Zone and is divided into four beds of decimetric scale. Three main microfacies are present. (1) oncoid rudstones, (2) ooid bioclastic packstones and (3) silty burrowed wackestones/packstones. Sedimentation took place in a very quiet environment, below the photic zone and below or near the storm wave base. The general setting is a distal carbonate ramp, its lower part characterized by hemipelagic sedimentation indicated by the presence of planktonic foraminifers. The inferred depth is around 100 m. Free oxygen concentration was low. Dysaerobic conditions are indicated by a scarcity of benthic macrofauna. Ferruginous structures are numerous in the first two microfacies, and absent in the last. Hematite staining is not uniform and follows many sedimentary patterns. Among the more widespread Fe structures are perforation infillings with endolithic microorganisms, microstromatolites, oncoids, ooids, blisters, coatings and hardgrounds. These structures can be associated and none are mutually exclusive. Hematite-coated filaments of different sizes and shapes are observed in the micrite matrix: the walls of various organisms; the calcite crystals associated with the Fe cortical laminations; the perforations and burrow; the hard-grounds; and microstromatolites. Petrographical and SEM examinations suggest that the laminated crusts (oncoids and hard-grounds) are formed by microbial iron mars dominated by filamentous bacteria and fungi. Seven types of microbes are recognized: filaments (five morphotypes), spheroidal bodies and stalked bodies. Filamentous microfossils of type 1 to 4 resemble the present-day filamentous bacteria (Beggiatoales and Cytophagaceae). Because of their large diameter and their branching nature, filaments of type 5 are possibly filamentous fungi. Another argument in favor of fungi is the presence of stalked and spheroidal bodies that resemble zoosporangia and oogonia of some Oomycota. In deep, calm and dysaerobic waters, many interfaces (e.g. between aerobic and dysaerobic waters) are present in the sediments. The stability of the soluble reduced state of iron (Fe2+) is higher at such interfaces, and many ferric iron-encrusted microbial fossils are observed. iron could thus serve as an electron donor for microbial iron-oxidation processes. Other microbial iron deposition pathways are also possible. It appears that, regardless of geological age (Paleozoic, Mesozoic) and geographical location, the same microbiological mechanisms an probably responsible for the red color in calcareous stratified or unstratified bodies. The presence of fossilized iron-encrusted bacteria and fungi at interfaces may therefore serve as an indicator of anoxic to dysaerobic conditions in various paleo(micro)environments. (C) 2000 Elsevier Science B.V. All

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Ponzoni S, Guimaraes FS, Del Bel EA, Garcia-Cairasco N. 2000. Behavioral effects of intra-nigral microinjections of manganese chloride: Interaction with nitric oxide. Progress in Neuro-Psychopharmacology & Biological Psychiatry 24(2):307-325.
Abstract: 1. Microinjection of manganese chloride (MnCl2) into the rat substantia nigra pars compacta (SNc) induces a neurodegenerative process manifested by apomorphine-induced rotational behavior. Manganese intoxication produces a parkinsonism-like phenotype in humans. 2. In addition to motor control the substantia nigra has also been proposed to be related to epilepsy and emotional behavior. 3. Although nitric oxide (NO) participation in neurodegenerative processes is still questioned, neurons stained for NAPDH-diaphorase, a marker of NO-producing cells, are spared in several experimental neuronal lesions. Additionally, NO has also been suggested to participate in motor control. 4. The objective of this study was to analyze the effects of MnCl2-induced nigral degeneration in audiogenic seizure susceptibility, anxiety and motor activity. We also analyzed if NO synthesis inhibition (N-G-nitro-L-arginine 25 mg/Kg twice a day for 4 days) modifies MnCl2 induced neurodegenerative process. 5. MnCl2 (50 mu g) microinjection into the SNc caused a statistical significant higher number of apomorphine (0.75 mg/kg s.c.)-induced rotations. No sensitization to audiogenic seizure was found but the lesion induced an increase of open arm exploration in the elevated plus maze, suggesting an anxiolytic effect. 6. The MnCl2-nigral lesion was accompanied by an increased number of NADPH-d positive neurons in the ipsilateral SNc and striatum (both sides). NO synthesis inhibition potentiated the MnCl2-nigral lesion and reversed the NADPH-d cell number increase. 7. The present results show that MnCl2-nigral lesion can influence emotional behavior and suggest that NO may modify the progression of manganese-induced degenerative process.

Pinero DJ, Li NQ, Connor JR, Beard JL. 2000. Variations in dietary iron alter brain iron metabolism in developing rats. J Nutr 130(2):254-263.
Abstract: The rat has been widely used as a model for the study of iron deficiency (ID), but the differences in the timing of development of humans and rats must be taken into account to derive appropriate conclusions from the animal model. This study was designed to evaluate the effects of dietary ID and iron excess on rat brain iron and the iron metabolism proteins, transferrin (Tf), transferrin receptor (TfR) and ferritin, The experimental design is developmentally sensitive and permits control of the timing as well as the duration of the nutritional insult. Iron-deficient and iron-supplemented (SU) rats between postnatal day (PND) 10 and 21, PND 21 and 35 and PND 10 and 35 were used to study the effects of early, late, and long-term iron deficiency and supplementation. Some ID rats were iron repleted between PND 21 and 35, These experiments demonstrated several new findings: 1) Early ID/SU (PND 10-21) altered brain iron, TFR, Tf and ferritin concentration in many regions different from those observed in the later period (PND 21-35). 2) Two weeks of iron repletion were adequate for correcting the overall Fe concentration of the brain and of individual brain regions, although larger amounts of iron were necessary to fully normalize iron and its regulatory proteins. 3) Long-term ID/SU resulted accordingly in the continued decrease or increase in brain iron concentration in some brain regions and not others. In conclusion, brain regions regulate their iron concentration in response to local needs when faced with alterations in systemic iron delivery.

Pinero DJ, Hu J, Connor JR. 2000. Alterations in the interaction between iron regulatory proteins and their iron responsive element in normal and Alzheimer's diseased brains. Cell Mol Biol 46(4):761-776.
Abstract: Iron regulatory proteins (IRPs) are cytoplasmic mRNA binding proteins involved in intracellular regulation of iron homeostasis. IRPs regulate expression of ferritin and transferrin receptor at the mRNA level by interacting with a conserved RNA structure termed the iron-responsive element (IRE). This concordant regulation of transferrin receptors and

ferritin is designed so a cell can obtain iron when it is needed, and sequester iron when it is in excess. However, we have reported that iron accumulates in the brain in Alzheimer's disease without a concomitant increase in ferritin. An increase in iron without proper sequestration can increase the vulnerability of cells to oxidative stress. Oxidative stress is a component of many neurological diseases including Alzheimer's. We hypothesized that alterations in the IRP/IRE interaction could be the site at which iron mismanagement occurs in the Alzheimer's brains. In this report we demonstrate that in normal human brain extracts, the IRP is detected as a double IRE/IRP complex by RNA band shift assay, but in 2 of 6 Alzheimer's brain (AD) extracts examined a single IRE/IRP complex was obtained. Furthermore, the mobility of the single IRE/IRP complex in Alzheimer's brain extracts is decreased relative to the double IRE/IRP complex. Western blot and RNA band super shift assay demonstrate that IRP1 is involved in the formation of the single IRE/IRP complex. in vitro analyses suggest that the stability of the doublet complex and single AD complex are different. The single complex from the AD brain are more stable. A more stable IRE/IRP complex in the AD brain could increase stability of the transferrin receptor mRNA and inhibit ferritin synthesis. At the cellular level, the outcome of this alteration in the molecular regulatory mechanism would be increased iron accumulation without an increase in ferritin; identical to the observation we reported in AD brains. The appearance of the single IRE/IRP complex in Alzheimer's brain extracts is associated with relatively high endogenous ribonuclease activity. We propose that elevated RNase activity is one mechanism by which the iron regulatory system becomes dysfunctional.

Pinero DJ, Connor JR. 2000. Iron in the brain: An important contributor in normal and diseased states. Neuroscientist 6(6):435-453.
Abstract: Iron is essential for normal neurological function because of its role in oxidative metabolism and because it is a cofactor in the synthesis of neurotransmitters and myelin. In the past several years, there has been increased attention to the importance of oxidative stress in the central nervous system. Iron is the most important inducer of reactive oxygen species, therefore, the relation of iron to neurodegenerative processes is more appreciated today than it was a few years ago. Nevertheless, despite this increased attention and awareness, our knowledge of iron metabolism in the brain at the cellular and molecular levels is still limited. Iron is distributed in a heterogeneous fashion among the different regions and cells of the brain. This regional and cellular heterogeneity is preserved across many species. Brain iron concentrations are not static; they increase with age and in many diseases and decrease when iron is deficient in the diet. In infants and children, insufficient iron in the diet is associated with decreased brain iron and with changes in behavior and cognitive functioning. Abnormal iron accumulation in the diseased brain areas and, in some cases, alterations in iron-related proteins have been reported in many neurodegenerative diseases, including Hallervorden-Spatz syndrome, Alzheimer's disease, Parkinson's disease, and Friedreich's ataxia. There is strong evidence for iron-mediated oxidative damage as a primary contributor to cell death in these disorders. Demyelinating diseases, such as multiple sclerosis, especially warrant study in relation to iron availability. Myelin synthesis and maintenance have a high iron requirement, thus, oligodendrocytes must have a relatively high and constant supply of iron. However, the high oxygen utilization, high density of lipids, and high iron content of white matter all combine to increase the risk of oxidative damage. We review here the current knowledge of the normal metabolism of iron in the brain and the suspected role of iron in neuropathology.

Paulson GW. 2000. Restless legs syndrome - How to provide symptom relief with drug and nondrug therapies. Geriatrics 55(4):35-38, 43-44, 47-48.
Abstract: Restless legs syndrome (RLS) is a perplexing, debilitating, and fairly common condition that can be challenging to manage. Hallmark symptoms include an increase in the severity of sensations during rest and an irresistible urge to move the affected limbs. RLS often occurs

concomitantly with periodic limb movement disorder, There are no known causes of RLS, but likely triggers include heredity, iron and vitamin deficiencies, caffeine, and alcohol. Chronic conditions such as diabetes, peripheral neuropathy, and Parkinson's disease can worsen and prolong RLS symptoms. Symptom management begins by establishing proper nutrition intake and improved sleep hygiene. If these fail, conservative pharmacologic treatment is appropriate, with regimens chosen from dopaminergic agents, benzodiazepines, opioids, and anticonvulsants.

Paik SR, Shin HJ, Lee JH. 2000. Metal-catalyzed oxidation of alpha-synuclein in the presence of copper(II) and hydrogen peroxide . Arch Biochem Biophys 378(2):269-277.
Abstract: alpha-Synuclein is a component of abnormal protein depositions of Lewy bodies and senile plaques found in Parkinson's and Alzheimer's diseases, respectively, By using chemical coupling reagents such as dicyclohexylcarbodiimide or N-(ethoxycarbonyl)-2-ethoxy-1,2-dihydroquinoline, the protein was shown to experience self-oligomerization in the presence of either copper(II) or A beta 25-35, The oligomers which appeared as a ladder on a 10-20% Tricine/SDS-PAGE have been suggested to participate in the formation of protein aggregations by possibly providing a nucleation center, Since oxidatively modified protein could increase its own tendency toward protein aggregation, metal-catalyzed oxidation of alpha-synuclein has been examined with copper(II) and hydrogen peroxide in the absence of the coupling reagent, intriguingly, the protein was also self-oligomerized into an SDS-resistant ladder on the gel. This biochemically specific copper-mediated oxidative oligomerization was shown to be dependent upon the acidic C-terminus of alpha-synuclein because the C-terminally truncated proteins such as alpha-syn114 and alpha-syn97 were not affected by the metal and hydrogen peroxide, More importantly, the oxidative oligomerization was synergistically enhanced by the presence of A beta 25-35, indicating that the peptide interaction with alpha-synuclein facilitated the copper(II) binding to the acidic C-terminus and subsequent oxidative crosslinking, It has been, therefore, suggested that abnormalities in copper and H2O2 homeostasis and certain pathological factors functionally similar to the A beta 25-35 could play critical roles in the metal-catalyzed oxidative oligomerization of alpha-synuclein, which may lead to possible protein aggregation and neurodegenerations, (C) 2000 Academic Press.

Ostrerova-Golts N, Petrucelli L, Hardy J, Lee JM, Farer M, Wolozin B. 2000. The A53T alpha-synuclein mutation increases iron-dependent aggregation and toxicity. J Neurosci 20(16):6048-6054.
Abstract: Parkinson's disease (PD) is the most common motor disorder affecting the elderly. PD is characterized by the formation of Lewy bodies and death of dopaminergic neurons. The mechanisms underlying PD are unknown, but the discoveries that mutations in alpha-synuclein can cause familial PD and that alpha-synuclein accumulates in Lewy bodies suggest that alpha-synuclein participates in the pathophysiology of PD. Using human BE-M17 neuroblastoma cells overexpressing wild-type, A53T, or A30P alpha-synuclein, we now show that iron and free radical generators, such as dopamine or hydrogen peroxide, stimulate the production of intracellular aggregates that contain alpha-synuclein and ubiquitin. The aggregates can be identified by immunocytochemistry, electron microscopy, or the histochemical stain thioflavine S. The amount of aggregation occurring in the cells is dependent on the amount of alpha-synuclein expressed and the type of alpha-synuclein expressed, with the amount of alpha-synuclein aggregation following a rank order of A53T > A30P > wild-type > untransfected. In addition to stimulating aggregate formation, alpha-synuclein also appears to induce toxicity. BE-M17 neuroblastoma cells overexpressing alpha-synuclein show up to a fourfold increase in vulnerability to toxicity induced by iron. The vulnerability follows the same rank order as for aggregation. These data raise the possibility that alpha-synuclein acts in concert with iron and dopamine to induce formation of Lewy body pathology in PD and cell death in PD.

Obata T, Yamanaka Y. 2000. Protective effect of histidine on potassium chloride depolarization enhances 1-methyl-4-phenylpyridinium ion-induced hydroxyl radical generation in the rat striatum. Life Sci 68(6):689-697.
Abstract: The present study examined the antioxidant effect of histidine on extracellular potassium ion concentration, [K+](o)-induced depolarization enhances 1-methyl-4-phenylpyridinium ion (MPP+)-induced hydroxyl radical (. OH) generation in the rat striatum. Rats were anesthetized and sodium salicylate in Ringer's solution (0.5 nmol/M muL/min) was infused through a microdialysis probe to detect the generation of . OH as reflected by the nonenzymatic formation of 2,3-dihydroxybenzoic acid (DHBA) in the striatum. Induction of [K+](o) (20, 70 and 140 mM) significantly increased the level of 2,3-DHBA by the action of MPP+ (5 mM) in a concentration-dependent manner. However, histidine (25 mM) reduced the [K+](o)-induced . OH formation. Although the level of MPP+-induced dopamine (DA) and 2,3-DHBA formation after [K+](o) (70 mM) treatment increased, [K+], failed to increase either the level of MPP+-induced DA and 2,3-DHBA in the reserpinized group. When iron (II) was administered to [K+](o) (70 mM)-pretreated rats, iron (II) clearly produced a dose-dependent increase in the level of 2,3-DHBA, as compared with MPP+-only treated rats. However, in the presence of histidine (25 mM), the effect of [K+](o) was abolished. These results indicated that histidine may reduce the [K+](o)-induced depolarization enhanced . OH formation by the action of MPP+ in the rat striatum. (C) 2000 Elsevier Science Inc. All rights reserved.

Obata T, Yamanaka Y. 2000. Methamphetamine enhances 1-methyl-4-phenylpyridinium ion-induced hydroxyl radical generation in the rat striatum. Neurosci Lett 292(1):54-56.
Abstract: We determined the methamphetamine (MA), a potent dopamine (DA) releaser, enhances 1-methyl-4-phenylpyridinium ion (MPP+)-induced hydroxyl radical (. OH) generation in the rat striatum. Rats were anesthetized, and sodium salicylate in Ringer's solution (0.5 nmol/mu l/min) was infused through a microdialysis probe to detect the generation of OH as reflected by the non-enzymatic formation of 2,3-dihydroxybenzoic acid (DHBA) in the striatum. After administration of MA (5 mg/kg i.v., every 2 h, four times), MA drastically increased DA release and the OH formation. When iron (II) was administered to the MA-treated animals, a marked elevation of DHBA was observed, compared with MPP+-only treated animals, that showed a positive linear correlation between DA and OH formation trapped as DHBA (R-2 = 0.985) in the dialysate. These results suggest that MA enhances the OH products of efflux/oxidation due to MPP+. (C) 2000 Published by Elsevier Science Ireland Ltd.

Obata T, Aomine M, Yamanaka Y. 2000. Potassium chloride depolarization enhances MPP+-induced hydroxyl radical generation in the rat striatum. Brain Res 852(2):488-491.
Abstract: We determined that extracellular potassium ion concentration, [K+](o)-induced depolarization, enhances 1-methyl-4-phenylpyridinium ion (MPP+)-induced hydroxyl radical (. OH) generation in the rat striatum. Rats were anesthetized, and sodium salicylate in Ringer's solution (0.5 nmol/mu l/min) was infused through a microdialysis probe to detect the generation of OH as reflected by the non-enzymatic formation of 2,3-dihydroxybenzoic acid (DHBA) in the striatum, Induction of high concentration KCl (70 mM) drastically increased formation of . OH trapped as DHBA by the action of MPP+. When dopamine (DA) was administered to the high KCl-treated animals, a marked elevation of DHBA was observed, compared with MPP+-only-treated animals, that showed a positive Linear correlation between DA and . OH formation trapped as DHBA (R-2 = 0.979) in the dialysate, When corresponding experiments were performed with iron (II), the same results were obtained: a positive linear correlation between the release of iron (II) and DHBA (R-2 = 0.988) in the dialysate. These results suggest that [K+](o)-induced depolarization enhances the formation of . OH products of efflux/oxidation due to MPP+. (C) 2000 Elsevier Science B.V. All rights reserved.

Newman PE. 2000. Alzheimer's disease revisited. Med Hypotheses 54(5):

774-776.
Abstract: In a previous paper, it was suggested that a relative deficiency of essential fatty acids might play a role in the etiology of sporadic or non-familial Alzheimer's disease. A recent article regarding dementia in the Rotterdam Study reinforces this suggestion. It is also hypothesized that this relative deficiency could facilitate passage of aluminum into the brain, aluminum being increasingly suggested as one of the possible pathogenic factors in AD. It is further suggested that hypomethylation caused by a deficiency of S-adenosylmethionine might also play a role in the etiology of this disease and perhaps even of Parkinson's disease. (C) 2000 Harcourt Publishers Ltd.

Neumann M, Adler S, Schluter O, Kremmer E, Benecke R, Kretzschmar HA. 2000. alpha-Synuclein accumulation in a case of neurodegeneration with brain iron accumulation type 1 (NBIA-1, formerly Hallervorden-Spatz syndrome) with widespread cortical and brainstem-type Lewy bodies. Acta Neuropathol (Berl) 100(5):568-574.
Abstract: We studied a 27-year-old woman who died after a 6-year history of progressive dementia, dystonia, ataxia. apraxia. spasticity. choreoathetosis, visual and auditory hallucinations, and optic atrophy. Magnetic resonance imaging showed decreased intensity in the globus pallidus. substantia nigra, and dentate nuclei in T2-weighted images. supporting the clinical diagnosis of neurodegeneration with brain iron accumulation type (NBIA-1: formerly known as Hallervorden-Spatz syndrome. At autopsy the brain showed mild frontotemporal atrophy and discoloration of the globus pallidus and the substantia nigra pars reticularis. Histologically, features typical of NBIA-1 were found including widespread axonal spheroids and large deposits of iron pigment in the discolored regions. Additionally, excessive numbers of Lewy bodies (LBs) were found throughout all examined brain stem and cortical regions. LBs of both types, as well as Lewy neurites in this case of NBIA-1. were strongly labeled by antibodies against alpha-synuclein. These findings give further evidence that accumulation of a-synuclein is generally associated with LB formation, i.e., in Parkinson's disease, dementia with Lewy bodies and NBIA-1. The case presented here is particularly notable for its high number of LBs in all areas of the cerebral cortex.

Nelson SR, Pazdernik TL, Samson FE. 2000. Measurement of loosely-bound iron in brain regions using redox cycling and salicylate . Cell Mol Biol 46(3): 649-655.
Abstract: A sensitive iron assay was developed for measuring non-heme and loosely bound iron in regions of rat brain. The method is based on the salicylate trapping of hydroxyl radicals generated from ascorbate-driven redox cycling of Fe3+-EDTA. This assay has high sensitivity (about 20 nM) because of amplification obtained with redoxcycling and fluorescent detection of the salicylate hydroxylation product, 2,5-dihydroxybenzoate. The assay detects iron as Fe2+ and Fe3+ combined. Values of non-heme and loosely bound iron are given for three areas of cortex, caudate, hippocampus, thalamus and brainstem of the rat brain.

Nappi AJ, Vass E. 2000. Iron, metalloenzymes and cytotoxic reactions. Cell Mol Biol 46(3):637-647.
Abstract: There is considerable evidence implicating iron and other redox-active transition metals as progenitors of reactive intermediates of oxygen (ROI), molecules which lead to oxidative stress and contribute to various neurodegenerative processes. An important aspect of such metal-mediated damage to biomolecules is the site-specific nature of such pathological activity. Iron sequestering molecules, such as ferritin, transferrin, lactotransferrin, melanotransferrin, hemosiderin and heme can serve as cytoprotectants against metal-mediated oxidant damage. Metalloenzymes also constitute an important group Of iron sequestering molecules. Metalloenzyme-catalyzed reactions in which metal ions at the enzyme active site undergo redox-cycling in association with O-2 are site-specific in nature, and may represent a potential source of ROT-mediated damage to biomolecules. Dysregulation of brain iron and alterations in the levels of

metalloenzymes involved in reactions with O-2 derived molecules can contribute to neuronal damage. Iron may increase the cytotoxicity of neuronal dopamine by increasing its rate of oxidation to quinones and semiquinones, thereby reducing the level of this neurotransmitter. Interestingly, dopamine also may play an important role in the maintenance of transition-metal homeostasis as an iron chelator, since it can form both catecholate and hydroxamate groups, molecules employed by many microorganisms to sequester iron.

Munch G, Luth HJ, Wong A, Arendt T, Hirsch E, Ravid R, Riederer P. 2000. Crosslinking of alpha-synuclein by advanced glycation endproducts - an early pathophysiological step in Lewy body formation? J Chem Neuroanat 20(3-4):253-257.
Abstract: An excess of reactive carbonyl compounds (carbonyl stress) and their reaction products, advanced glycation endproducts (AGEs), are thought to play a decisive role in the pathogenesis of neurodegenerative disorders and Parkinson's disease (PD) in particular. Accumulation of AGEs in various intracellular pathological hallmarks of PD. such as Lewy bodies, densely crosslinked intracellular protein deposits formed from neurofilament components and alpha -synuclein, have already been described in patients in advanced stages of the disease. There is, however, no indication of the involvement of AGE-induced crosslinking of alpha -synuclein in very early stages of the disease. In this study, we observed that AGEs and alpha -synuclein are similarly distributed in very early Lewy bodies in the human brain in cases with incidental Lewy body disease. These cases might be viewed as pre-Parkinson patients, i.e. patients who came for autopsy before the possible development of clinical signs of PD. AGEs are both markers of transition metal induced oxidative stress as well as, inducers of protein crosslinking and free radical formation by chemical and cellular processes. Thus, it is likely that AGE promoted formation of Lewy bodies reflects very early causative changes rather than late epiphenomenons of PD. (C) 2000 Elsevier Science B.V. All rights reserved.

Mori N, Hirayama K. 2000. Long-term consumption of a methionine-supplemented diet increases iron and lipid peroxide levels in rat liver. J Nutr 130(9): 2349-2355.
Abstract: Methionine is a protective factor against various types of liver damage, but excessive dietary methionine is hepatotoxic. Because the mechanisms of L-methionine-related hepatotoxicity are poorly understood, the effect of long-term excessive L-methionine intake on the metabolism of iron and antioxidants was studied in rat liver to determine whether oxidative stress is involved. Wistar male rats were fed either an L-methionine-supplemented (16.0 g/kg) diet or a control diet for 1, 3, 6 and 9 mo. The growth rate of L-methionine-supplemented rats was significantly slower than that of controls. Iron, ferritin and thiobarbituric acid-reactive substances (TBARS) levels in the liver were greater in supplemented rats than in controls. Serum iron and transferrin levels were significantly lower in L-methionine-treated rats compared with controls. Serum ferritin did not differ between the two groups. Hepatic glutathione peroxidase activity, catalase activity and total glutathione concentrations were higher in rats fed the L-methionine-supplemented diet at 1 and 3 mo, but not at 6 and 9 mo. These results indicate that long-term consumption of excess L-methionine by rats may affect primarily iron metabolism rather than the antioxidant defense system and, consequently, induce an accumulation of iron.

Moos T, Trinder D, Morgan EH. 2000. Cellular distribution of ferric iron, ferritin, transferrin and divalent metal transporter 1 (DMT1) in substantia nigra and basal ganglia of normal and beta 2-microglobulin deficient mouse brain. Cell Mol Biol 46(3):549-561.
Abstract: We examined whether high levels of circulatory iron may cause iron accumulation in the brain. In particular, we focussed on the substantia nigra and basal ganglia as several papers have indicated that iron may accumulate here and cause death of dopaminergic neurons. Normal mice and a mouse model of hereditary haemochromatosis, the beta 2-

microglobulin (beta 2m) knock out [beta 2m (-/-)] mouse, which has high levels of circulating iron due to increased iron absorption, were examined. The iron concentration in livers were: 170 +/- 15 mu g/g (mean +/- SD) in controls and 1010 +/- 50 mu g/g in beta 2m (-/-) mice ($p<0.001$), whereas in the brain the respective values were 47 +/- 1 mu g/g and 53 +/- 2 mu g/g ($p<0.02$). Hence, the difference between cerebral iron levels of normal and beta 2m (-/-) mice was small. Histological examination of the brains revealed an unequivocal distribution of ferric iron, ferritin, transferrin and divalent metal transporter 1 (DMT1), which were indistinguishable when normal and beta 2m (-/-) mice were compared. In the substantia nigra and basal ganglia, ferric iron and the iron-binding proteins were present in identical cell types, which mainly comprised oligodendrocytes and microglia. Neurons were lightly labelled with transferrin and DMT1. The virtual lack of an increase in cerebral iron in beta 2m (-/-) mice clearly shows that the blood-brain barrier (BBB) is capable of restricting the transport of excess plasma iron into the brain.

Montine TJ, Amarnath V, Picklo MJ, Sidell KR, Zhang J, Graham DG. 2000. Dopamine mercapturate can augment dopaminergic neurodegeneration. Drug Metab Rev 32(3-4):363-376.
Abstract: Pathological and biochemical studies have consistently associated endogenous catechol oxidation with dopaminergic neurodegeneration in Parkinson's disease (PD). Recently, it has been proposed that products of catechol oxidation, the catechol thioethers, may contribute to dopaminergic neurodegeneration. In other organ systems, thioether cytotoxicity is influenced profoundly by the mercapturic acid pathway. We have pursued the hypothesis that endogenous catechol thioethers produced in the mercapturic acid pathway contribute to dopaminergic neurodegeneration. Our results showed that the extent of in vitro metal-catalyzed oxidative damage by catechol thioethers varied with the structures of the parent catechol and thioether adduct. Catechol mercapturates uniquely produced more oxidative damage than their parent catechols. In dopaminergic cell cultures, dopamine induced apoptosis in a concentration-dependent manner from 5 to 50 muM. The apoptotic effect of dopamine was greatly enhanced by subcytotoxic concentrations of the mitochondrial inhibitor, N-methyl-4-phenylpyridinium (MPP+). similarly. subcytotoxic levels of the mercapturate or homocysteine conjugate of dopamine significantly augmented dopamine-induced apoptosis. Finally, microsomal fractions of substantia nigra from PD patients or age-matched controls had comparable cysteine-S-conjugate N-acetyltransferase activity. These data indicate that the mercapturate conjugate of dopamine may augment dopaminergic neurodegeneration and that the mercapturate pathway exists in human substantia nigra.

Miyazaki I, Sogawa CA, Asanuma M, Higashi Y, Tanaka K, Nakanishi T, Ogawa N. 2000. Expression of metallothionein-III mRNA and its regulation by levodopa in the basal ganglia of hemi-parkinsonian rats. Neurosci Lett 293 (1):65-68.
Abstract: In the brain, metallothionein (MT)-III exhibits a free radical scavenging activity. Here we examined the expression of MT-III mRNA in the basal ganglia of 6-hydroxydopamine (6-OHDA)-lesioned hemi-parkinsonian rats and its regulation by levodopa. The level of MT-III mRNA was significantly decreased in the striatum of 6-OHDA-lesioned side. Levodopa treatment significantly increased the expression of striatal MT-III mRNA in the non-lesioned side, but showed no significant effect in the 6-OHDA-lesioned side. These results suggest that the regulation of MT-III mRNA may be related to the progressive degeneration in parkinsonism. (C) 2000 Published by Elsevier Science Ireland Ltd.

Miura T, Muraoka S, Fujimoto Y, Zhao KC. 2000. DNA damage induced by catechol derivatives. Chem Biol Interact 126(2):125-136.
Abstract: We investigated the effect of catechol derivatives, including dopa, dopamine, adrenaline and noradrenaline, on DNA damage and the mechanisms of DNA strand breakage and formation of 8-hydroxyguanine (8HOG). The catechol derivatives caused strand breakage of plasmid DNA

in the presence of ADP-Fe3+. The DNA damage was prevented by catalase, mannitol and dimethylsulfoxide, suggesting hydroxyl radical (HO..)-like species are involved in the strand breakage of DNA. Iron chelators, such as desferrioxamine and bathophenanthroline, and reduced glutathione also inhibited the DNA damage. Deoxyribose, a molecule that is used to detect HO., was not degraded by dopa in the presence of ADP-Fe3+. By adding EDTA, however, dopa induced the marked deoxyribose degradation in the presence of ADP-Fe3+, indicating that EDTA may extract iron from ADP-Fe3 + to catalyze HO. formation by dopa. Thus, EDTA was a good catalyst for HO.-generation, whereas it did not promote the strand breakage of DNA. However, calf thymus DNA base damage, which was detected as s-HOG formation, was caused by dopa in the presence of EDTA-Fe3+, but not in the presence of ADP-Fe3+. The 8HOG formation was also inhibited by catalase and HO. scavengers, indicating that HO. was involved in the base damage. These results suggest that DNA strand breakage is due to ferryl species rather than HO., and that 8HOG formation is due to HO. rather than ferryl species. (C) 2000 Published by Elsevier Science Ireland Ltd. All rights reserved.

Missy P, Lanhers MC, Grignon Y, Joyeux M, Burnel D. 2000. In vitro and in vivo studies on chelation of manganese. Human & Experimental Toxicology 19 (8):448-456.
Abstract: This work deals with new chelating agents of manganese (Mn). Out of 24 compounds chosen for their chemical structure supposed to be Favorable for Mn complexation. six polyaminopolycarboxylic acids proved to be efficient for displacing Mn bound to serum bovine proteins in vitro: TTHA. DTPA. DPTA, DPTA-OH. HEED. EDTA (mobilization greater than or equal to 50%). The first Five compounds were then tested in vivo on rats pre treated with MnCl2. They exhibited only slight to moderate efficacy to diminish Mn in tissues and were ineffective on increased hin concentration in whole blood: in addition, they had different and specific mobilizing effects on other essential elements (Fe. Zn. Cu). Their limited efficacy in vivo could be due to the formation of very stable complexes between Mn2 + and different molecules such as hemoglobin and certain cytochromes, instead of Fe2+. This could disturb the functioning of the cellular respirator?; chain. leading to an incomplete reduction of O-2 with formation of free oxygenated radicals, reduction in the energy supply, and disturbance of the cytochromes renewal mechanism. All of these phenomena could accelerate cellular aging and explain the lack of efficacy of the chelating agents towards Mn neurotoxicity (Parkinson's syndrome).

Mirza B, Hadberg H, Thomsen P, Moos T. 2000. The absence of reactive astrocytosis is indicative of a unique inflammatory process in Parkinson's disease. Neuroscience 95(2):425-432.
Abstract: Virtually any neurological disorder leads to activation of resident microglia and invasion of blood-borne macrophages, which are accompanied by an increase in number and change in phenotype of astrocytes, a phenomenon generally termed reactive astrocytosis. One of the functions attributed to activation of astrocytes is thought to involve restoration of tissue damage. Hitherto, the role of astrocytes in the inflammatory reaction occurring in Parkinson's disease has not received much attention. In the present study, we examined the inflammatory events in autopsies of the substantia nigra and putamen from Parkinson's disease patients using age-matched autopsies from normal patients as controls. In the substantia nigra, activation of microglia was consistently observed in all Parkinson's disease autopsies as verified from immunohistochemical detection of CR3/43 and ferritin. Activation of resident microglia was not observed in the putamen. No differences were observed between controls and Parkinson's disease autopsies from the substantia nigra and putamen, in terms of distribution, cellular density or cellular morphology of astrocytes stained for glial fibrillary acidic protein or metallothioneins I and II, the latter sharing high affinity for metal ions and known to be induced in reactive astrocytes, possibly to exert anti-oxidative effects. Together, these findings indicate that the inflammatory process in Parkinson's disease is characterized by activation of resident microglia

without reactive astrocytosis, suggesting that the progressive loss of dopaminergic neurons in Parkinson's disease is an ongoing neurodegenerative process with a minimum of involvement of the surrounding nervous tissue. The absence of reactive astrocytosis in Parkinson's disease contrasts what follows in virtually any other neurological disorder and may indicate that the inflammatory process in Parkinson's disease is a unique phenomenon. (C) 1999 IBRO. Published by Elsevier Science Ltd.

Lopiano L, Chiesa M, Digilio G, Giraudo S, Bergamasco B, Torre E, Fasano M. 2000. Q-band EPR investigations of neuromelanin in control and Parkinson's disease patients. Biochimica Et Biophysica Acta-Molecular Basis of Disease 1500(3):306-312.
Abstract: New insights into the understanding of the changes induced in the iron domain of neuromelanin (NM) upon development of Parkinson's disease (PD) have been gained by electron paramagnetic spectroscopy (EPR). The results of this study are compared with a previously reported variable temperature analysis of X-band EPR spectra of a NM specimen obtained from control brain tissues. The availability of high sensitivity instruments operating in the Q-band (34.4 GHz) allows us to deal with the low amounts of NM available from PD brains. The organization of iron in NM is in the form of polynuclear superparamagnetic/antiferromagnetic aggregates, but the lack of one or more signals in the EPR spectra of NM from PD suggests that the development of the pathology causes NM to decrease its ability to bind iron. Furthermore, the detection of the Mn(II) signal in the Q-band spectra is exploited as an additional internal probe to assess minor structural differences in iron domains of PD and control NM specimens. (C) 2000 Published by Elsevier Science B.V. All rights reserved.

Liu HM, Tsai SJJ, Cheng FC, Chung SY. 2000. Determination of trace manganese in the brain of mice subjected to manganese deposition by graphite furnace atomic absorption spectrometry. Anal Chim Acta 405(1-2): 197-203.
Abstract: Trace manganese determination in mouse brain tissues by graphite furnace atomic absorption spectroscopy with a deuterium lamp background corrector and Ca(OH)(2) as modifier was described. The manganese content was investigated in four brain locations (cortex, hippocampus, substantia nigra, striatum) in mice subjected to single or multiple injections of manganese chloride. Substantia nigra tissues revealed the highest manganese content 24 h following single injection of manganese chloride. Large accumulations of manganese were observed within substantia nigra and striatum of mice subjected to four consecutive injections of manganese chloride. Thus, substantia nigra and striatum appear to be the brain regions most affected by manganese poisoning. The present method provided rapid and sensitive analysis of brain manganese. The detection limit (3 sigma) was 1.04 pg and the linearity ranged from 4 to 80 pg. With this method, manganese recoveries from mouse brain tissues ranged from 90-110%, and a complete analysis could be performed within 2 min. The preparation of brain homogenization samples is universal, and samples are less susceptible to contamination. Therefore, other neurochemical substances can be analyzed in remaining samples. (C) 2000 Elsevier Science B.V. All rights reserved.

Lingor P, Unsicker K, Krieglstein K. 2000. GDNF and NT-4 protect midbrain dopaminergic neurons from toxic damage by iron and nitric oxide. Exp Neurol 163(1):55-62.
Abstract: Free radical formation is considered to be a major cause of dopaminergic (DAergic) cell death in the substantia nigra leading to Parkinson's disease (PD). In this study we employed several radical donors including iron and sodium nitroprusside to induce toxic effects on DAergic neurons cultured from the embryonic rat midbrain floor. Overall cell survival was assessed by assaying LDH, and DAergic neuron survival was monitored by counting tyrosine hydroxylase-positive cells. Our data suggest that the DAergic neuron population is about fourfold more susceptible to free-radical-mediated damage than the total population of

midbrain neurons. Application of the neurotrophic factors GDNF and NT-4, for which DAergic neurons have specific receptors, prior to toxin administration protected these neurons from toxin-mediated death, which, fully or in part, occurs under the signs of apoptosis. These findings underscore the importance of GDNF and NT-4 in designing future therapeutical concepts for PD. (C) 2000 Academic Press.

Linert W, Jameson GNL. 2000. Redox reactions of neurotransmitters possibly involved in the progression of Parkinson's Disease. J Inorg Biochem 79 (1-4):319-326.
Abstract: In Parkinson's Disease the neuromelanin in the substania nigra is known to contain considerably increased amounts of iron suggesting the presence of free, unprotected iron ions during its formation. Iron(II) is known to interact with peroxide via Fenton's reaction producing OH-radicals or ferryl (Fe(IV)) species. This can readily oxidize the neurotransmitter dopamine to the neurotoxic 6-hydroxydopamine (6-OHDA) which is a strong reducing agent. The produced 6-OHDA is, in turn, able to reduce and possibly release iron, as iron(II), from the iron storage protein ferritin. This cycle of events could well explain the development of Parkinson's Disease due to a continuous production of cell damaging species. The contrasting behaviour of 6-OHDA with some other important catecholamines is discussed. (C) 2000 Elsevier Science Inc. All rights reserved.

Lin AMY, Ho LT. 2000. Melatonin suppresses iron-induced neurodegeneration in rat brain. Free Radic Biol Med 28(6):904-911.
Abstract: The antioxidative action of melatonin on iron-induced neurodegeneration in the nigrostriatal dopaminergic system was evaluated in vivo. Intranigral infusion of iron chronically degenerated the dopaminergic transmission of the nigrostriatal system. An increase in lipid peroxidation in the infused substantia nigra and reductions in dopamine levels and dopaminergic terminals in the ipsilateral striatum were observed 7 d after iron infusion, Whereas local infusion of melatonin (60 mu g/mu l, 1 mu l) alone did not alter dopaminergic transmission, coinfusion of melatonin with iron suppressed iron-induced oxidative damages. Systemic infusion of melatonin via osmotic pumps had no effect on iron-induced neurodegeneration. However, repetitive intraperitoneal injections of melatonin (10 mg/kg) prevented iron-induced oxidative injuries. The ratio of glutathione (GSH)/oxidized glutathione (GSSG) was moderately increased in the lesioned substantia nigra of the melatonin-treated rats compared to that of the lesioned group in control rats. The antioxidative effect of melatonin was verified in cortical homogenates. Melatonin dose-dependently suppressed autoxidation and iron-induced lipid peroxidation. Melatonin was as effective as GSH and was less effective than Trolox (a water-soluble analogue of vitamin E) in inhibiting iron-elevated lipid peroxidation of brain homogenates. Our data suggest that melatonin is capable of at least partially preventing the iron-induced neurodegeneration in the nigrostriatal dopaminergic system. (C) 2000 Elsevier Science Inc.

Lee PL, Halloran C, Cross AR, Beutler E. 2000. NADH-ferric reductase activity associated with dihydropteridine reductase. Biochem Biophys Res Commun 271(3):788-795.
Abstract: In mammals dietary ferric iron is reduced to ferrous iron for more efficient absorption by the intestine. Analysis of a pig duodenal membrane fraction revealed two NADH-dependent ferric reductase activities, one associated with a b-type cytochrome and the other not. Purification and characterization of the non-cytochrome ferric reductase identified a 31 kDa protein. MALDI-MS analysis and amino acid sequencing identified the ferric reductase as being related to the 26 kDa liver NADH-dependent quinoid dihydropteridine reductase (DHPR). The NADH-dependent DHPR ferric reductase activity was found to be pteridine independent since exhaustive dialysis did not reduce activity and heat-inactivation destroyed activity. Pn intestinal Caco-2 cells, DHPR mRNA levels were found to be regulated by iron. Thus, DHPR appears to be a dual function enzyme, a NADH-dependent dihydopteridine reductase and an

iron-regulated, NADH-dependent, pteridine-independent ferric reductase. (C) 2000 Academic Press.

Lee JW. 2000. Manganese intoxication. Arch Neurol 57(4):597-599.
Abstract: Manganese plays an important role as a cofactor in many enzymatic reactions in humans but in excess amounts can cause irreversible nervous system damage.(1,2) Although manganism is a rare condition, it can be the cause of complex nervous system symptoms, especially in the setting of environmental exposure.(3,4) Specifically, manganese is a well-known cause of dystonic parkinsonism.(5) This article highlights several historical descriptions of the clinical manifestations, pathological changes, and attempted therapeutic intervention in manganese intoxication.

Lee HS, Park CW, Kim YS. 2000. MPP+ increases the vulnerability to oxidative stress rather than directly mediating oxidative damage in human neuroblastoma cells. Exp Neurol 165(1):164-171.
Abstract: MPP+, an active metabolite of MPTP, causes a dopaminergic neuronal degeneration similar to that observed in Parkinson's disease. Current data suggest that MPP+-induced cytotoxicity may be mediated by oxygen free radicals. To evaluate this hypothesis, we first investigated whether MPP+ could cause oxidative stress by producing oxygen free radicals in the SH-SY5Y, human neuroblastoma cell line. MPP+ was toxic to the cells dose-dependently but did not increase the level of lipid peroxidation at toxic concentrations. Second, we examined the effects of various antioxidants and an inhibitor of nitric oxide synthase (NOS) on the development of MPP+ cytotoxicity. Pretreatment with antioxidants such as ascorbic acid, Trolox, phenyl-tertiary-butyl-nitrone (PBN), which show protective effects on tert-butyl hydroperoxide (tBOOH) toxicity did not attenuate MPP+ cytotoxicity. Similarly, the combination of antioxidant enzymes, SOD and catalase (50 U/ml, respectively), did not protect the cells from the toxic action of MPP+. Also N-nitro-L-arginine methyl ester (NAME), a competitive inhibitor of NOS, and combined incubation with NAME and antioxidant enzymes failed to attenuate MPP+ cytotoxicity. On the other hand, a sublethal dose of MPP+ potentiated iron and H2O2-induced cytotoxicity. These results suggest that oxygen free radicals may not be a primary cause of MPP+-induced cell death but that MPP+ increases the vulnerability of cells to oxidative stress. (C) 2000 Academic Press.

Laitinen LV. 2000. Leksell's unpublished pallidotomies of 1958-1962. Stereotact Funct Neurosurg 74(1):1-10.
Abstract: Lars Leksell performed posteroventral pallidotomies in 123 parkinsonian patients from 1958 through 1962. This previously unpublished series was analyzed by the present author 40 years later. The analysis was based on a number of old radiographs that were found accidentally as well as the available hospital notes. After pallidotomy, Leksell had left metal markers in the brain to illustrate the lesion site. Superimposition of the markers on modern MRI scans of patients with similar-sized heads shows a high degree of accuracy of Leksell's pallidotomy. Copyright (C) 2000 S. Karger AG, Basel.

Lai JCK, Minski MJ, Chan AWK, Lim L. 2000. Interrelations Between Manganese and Other Metal Ions in Health and DiseaseVolume 37. p 123-156. Metal Ions in Biological Systems, Vol 37: Metal Ions in Biological Systems.

Kim NH, Park SJ, Jin JK, Kwon MS, Choi EK, Carp RI, Kim YS. 2000. Increased ferric iron content and iron-induced oxidative stress in the brains of scrapie-infected mice. Brain Res 884(1-2):98-103.
Abstract: Scrapie is a transmissible neurodegenerative disease of sheep and goats. The neuropathological changes include vacuolation, astrocytosis, the development of amyloid plaques in some instances, and neuronal loss. The mechanisms involved in neuronal cell death in scrapie are not known. Recently, we reported the presence of oxidative stress in the brains of scrapie-infected animals and suggested that this is the main

mechanism that induces neuronal cell loss. It is known that oxidative stress induced by free radicals is associated with iron accumulation; this association led to an examination of the levels of iron (total iron, Fe2+ and Fe3+) in the brains of control and scrapie-infected mice by biochemical methods. In the scrapie-infected group, both the level of total iron and the Fe3+ level were significantly increased in cerebral cortex, striatum, and brainstem as compared to the values in the control group. A shift in the ratio of Fe2+/Fe3+ was observed in the same regions of infected mice. Additionally, in this scrapie model, we confirmed the presence of oxidative stress, as evidenced by the increase of free malondialdehyde. These results suggest that iron metabolism is changed and that iron-induced oxidative stress partly contributes to neurodegeneration in scrapie infection. (C) 2000 Elsevier Science B.V. All rights reserved.

Khaldy H, Escames G, Leon J, Vives F, Luna JD, Acuna-Castroviejo D. 2000. Comparative effects of melatonin, L-deprenyl, Trolox and ascorbate in the suppression of hydroxyl radical formation during dopamine autoxidation in vitro. J Pineal Res 29(2):100-107.
Abstract: Degeneration of nigrostriatal dopaminergic neurons is the major pathogenic substrate of Parkinson's disease (PD). Inhibitors of monoamine oxidase B (MAO-B) have been used in the treatment of PD and at least one of them, i.e., deprenyl, also displays antioxidant activity. Dopamine (DA) autoxidation produces reactive oxygen species implicated in the loss of dopaminergic neurons in the nigrostriatal pathway. In this study we compared the effects of melatonin with those of deprenyl and vitamins E and C in preventing the hydroxyl radical (. OH) generation during DA oxidation. The rate of production of 2,3-dihydroxybenzoate (2,3-DHBA) in the presence of salicylate, an . OH scavenger, was used to detect the in vitro generation of . OH during iron-catalyzed oxidation of DA. The results showed a dose-dependent effect of melatonin, deprenyl and vitamin E in counteracting DA autoxidation, whereas vitamin C had no effect. Comparative analyses between the effect of these antioxidants showed that the protective effect of melatonin against DA autoxidation was significantly higher than that of the other compounds tested. Also, when melatonin plus deprenyl were added to the incubation medium, a potentiation of the antioxidant effect was found. These findings suggest that antioxidants may be useful in brain protection against toxicity of reactive oxygen species produced during DA oxidation, and melatonin, alone or in combination with deprenyl, may be an important component of the brain's antioxidant defenses to protect it from dopaminergic neurodegeneration.

Kaiser HE, Bodey B, Bodey B. 2000. Importance of treatment of depression in assuring the most efficacious management of Parkinson's disease. In Vivo 14(3):457-462.
Abstract: Parkinson's disease (PD) is characterized by pathological changes which include degeneration of dopaminergic neurons in the substantia nigra pars compacta coupled with intracytoplasmic inclusions known as Lewy bodies. Neurodegeneration and Lewy bodies can also be found in the locus coeruleus, nucleus basalis, hypothalamus, cerebral cortex, cranial nerve motor nuclei, and central and peripheral components of the autonomic nervous system. PD progression is associated with the development of dementia, autonomic dysfunction, and postural instability, which do not respond well to conventional therapy. Therapeutic efforts aimed at preventing or at least delaying PD progression by reducing the overload of iron and generation of ROS, correcting the zinc deficiency may be of great benefit. Current pharmacotherapy of PD, in addition to symptomatic L-dopa treatment, includes the neuroprotective strategies with dopamine agonists, monoamine oxidase-B inhibitors (MAO-B), glutamate antagonists, catechol O-methyltransferase (COMT) inhibitors and other antioxidants or free radical scavengers. Depression, anxiety disorder and stress are all associated with PD and it is therefore necessary to include treatment regimens for these ailments in addition to the traditional pharmacotherapy for the symptoms of PD, as well as the neuroprotective measures noted

above, in order to ensure the greatest possible benefit to PD patients.

Johnson S. 2000. Iron catalyzed oxidative damage, in spite of normal ferritin and transferrin saturation levels and its possible role in Werner's syndrome, Parkinson's disease, cancer, gout, rheumatoid arthritis, etc. Med Hypotheses 55(3):242-244.
Abstract: Iron loading in hemochromatosis attains extremely high levels and is accompanied by many signs (ferritin >300 mu g/l, hematocrit >50%, transferrin saturation >70%, etc.). Nevertheless, the disease is often overlooked by physicians, until several organs have been damaged permanently (heart, liver, brain, pancreas, kidneys, spleen, etc.). Therefore, severe oxidative damage catalyzed by Fe could occur, without the extremely high ferritin, hematocrit and transferrin saturation levels of hemochromatosis, and it is unlikely that it would ever be detected or even suspected. 1 postulate a mechanism, by which a cell can continue to express transferrin receptors, without producing ferritin, even when it is saturated with iron. Furthermore, I suggest that this silent iron loading, induced by cadmium and other metals, plays an important role in many degenerative diseases involving free radicals, DNA damage and peroxynitrite, all of which are intimately linked to iron. Moreover, since ferritin, transferrin saturation and hematocrit levels are not directly related to cellular iron levels, and since excess iron can wreak havoc in the cell, we can conclude that there is a need for a better way to evaluate intracellular iron levels and especially the intracellular free iron levels by a non-invasive technique. Finally, phlebotomy is suggested as the best way to reduce Fe and Mo stores, and chelation with succimer is recommended in order to eliminate Cd. (C) 2000 Harcourt Publishers Ltd.

Hori H, Ohmori O, Shinkai T, Kojima H, Okano C, Suzuki T, Nakamura J. 2000. Manganese superoxide dismutase gene polymorphism and schizophrenia: Relation to tardive dyskinesia. Neuropsychopharmacology 23(2):170-177.
Abstract: There has been increasing evidence that deranged superoxide dismutase (SOD) activities might be a risk factor for schizophrenia and/or tardive dyskinesia (TD). In the present study, we investiogated the genetic association between a functional polymorphism (Ala-9Val) in the human manganese (Mn) SOD gene and schizophrenia or TD (192 schizophrenics : 39 with TD and 153 without TD; 141 controls). No significant differences in the allelic or genotypic distribution between schizophrenics and controls were observed. However, we did find a significant difference in genotypic distribution between schizophrenics with and those without TD (p = .03). Moreover, decreased -9Ala (mutant) allele was found among patients with TD (p = .02; odds ratio = 0.29; 95% confidence interval = 0.10-0.83). In conjunction with previous findings of increased free radicals and decreased SOD activities in TD subjects, these results suggest that the -9Ala (high activity) MnSOD allele may play a role in protecting against susceptibility to TD in schizophrenics. (C) 2000 American College of Neuropsychopharmacology. Published by Elsevier Science Inc. All rights reserved.

Hellenbrand W, Bauer G, Boeing H, Seidler A, Robra BP. 2000. Diet in residents of East and West Germany in 1991-1992 as ascertained by a retrospective food frequency questionnaire. Soz Praventivmed 45(1):13-24.
Abstract: In this study, we compared dietary habits of residents in East (N = 76) and West Germany (N = 266) using results obtained in 1992-1993 from a retrospective semi-quantitative food frequency questionnaire referring to 1991 - 1992. Nutrient intakes were calculated based: on the German Federal Food Code. Univariate and multivariate logistic regression was used to determine whether dietary intakes varied according to residence in East and West Germany. At the food level, East German subjects reported a higher consumption of bread, spreadable fat, and sausage, whereas West German participants reported a higher intake of fruit, vegetables, and pasta and rice. At the macronutrient level, energy intake did not differ significantly between groups nor did the percent contribution of protein, carbohydrate, fat, and alcohol to total energy intake. East German participants had a lower total water and fibre intake,

the latter significant only after adjustment for confounders. At the micronutrient level, East German participants had a higher intake of cobalamin, retinol and retinol-equivalents (but not of beta-carotene). There were no differences in the intake of vitamins C, D, and E between groups Less salt and more potassium, calcium, magnesium and zinc were consumed by West than by East German subjects. Overall, both groups showed disadvantageous dietary patterns. The results are discussed in the context of an overview of other dietary surveys performed in the two parts of Germany before and after reunification. In general our results are consistent with other observations showing that dietary habits in East Germany rapidly approached those in West Germany after reunification, although some residual differences seemed to persist.

Ham D, Schipper HM. 2000. Heme oxygenase-1 induction and mitochondrial iron sequestration in astroglia exposed to amyloid peptides. Cell Mol Biol 46(3): 587-596.
Abstract: The mechanisms responsible for pathological iron deposition and mitochondrial insufficiency that have been documented in the brains of Alzheimer (AD) patients remain poorly understood. In the present study, we demonstrate that low-micromolar concentrations of amyloid(1-40) (A40) and amyloid(1-42) (A42), peptides implicated in the pathogenesis of AD, increase levels of heme oxygenase-1 (HO-1) mRNA and protein in cultured rat astroglia, Furthermore, 6 days of exposure to amyloid augments the sequestration of (FeCl3)-Fe-55-derived iron by astroglial mitochondria without affecting the disposition of this metal in whole-cell and lysosomal compartments. Mitochondrial iron deposition was not observed in the amyloid-treated glia when diferric-transferrin served as the metal donor. We had previously shown that inhibitors of HO-1 and the mitochondrial permeability transition pore (MTP) block the uptake of mitochondrial iron in astrocytes exposed to the pro-oxidant effects of dopamine and several pro-inflammatory cytokines. Similarly, in the current study, amyloid-induced mitochondrial iron trapping was significantly attenuated by co-administration of the HO-1 transcriptional suppressor, dexamethasone (DEX) or the MTP blocker, cyclosporin A (CSA). Thus, the marked enhancement of HO-1 expression previously demonstrated in AD-affected neurons and astroglia may transduce amyloid (oxidative) stress into the abnormal patterns of iron deposition and mitochondrial insufficiency characteristic of this disease. Finally, in experiments employing cytotoxic concentrations of A40, we provide evidence that inhibition of HO-1 transcription and related mitochondrial iron deposition may be an important mechanism by which DEX protects tissues subjected to amyloid stress.

Guo ZH, Ersoz A, Butterfield DA, Mattson MP. 2000. Beneficial effects of dietary restriction on cerebral cortical synaptic terminals: Preservation of glucose and glutamate transport and mitochondrial function after exposure to amyloid beta-peptide, iron, and 3-nitropropionic acid. J Neurochem 75(1): 314-320.
Abstract: Recent studies have shown that rats and mice maintained on a dietary restriction (DR) regimen exhibit increased resistance of neurons to excitotoxic, oxidative, and metabolic insults in experimental models of Alzheimer's, Parkinson's, and Huntington's diseases and stroke. Because synaptic terminals are sites where the neurodegenerative process may begin in such neurodegenerative disorders, we determined the effects of DR on synaptic homeostasis and vulnerability to oxidative and metabolic insults. Basal levels of glucose uptake were similar in cerebral cortical synaptosomes from rats maintained on DR for 3 months compared with synaptosomes from rats fed ad libitum. Exposure of synaptosomes to oxidative insults (amyloid beta-peptide and Fe2+) and a metabolic insult (the mitochondrial toxin 3-nitropropionic acid) resulted in decreased levels of glucose uptake. Impairment of glucose uptake following oxidative and metabolic insults was significantly attenuated in synaptosomes from rats maintained on DR. DR was also effective in protecting synaptosomes against oxidative and metabolic impairment of glutamate uptake. Loss of mitochondrial function caused by oxidative and metabolic insults, as

indicated by increased levels of reactive oxygen species and decreased transmembrane potential, was significantly attenuated in synaptosomes from rats maintained on DR. Levels of the stress proteins HSP-70 and GRP-78 were increased in synaptosomes from DR rats, consistent with previous data suggesting that the neuroprotective mechanism of DR involves a "preconditioning" effect. Collectively, our data provide the first evidence that DR can alter synaptic homeostasis in a manner that enhances the ability of synapses to withstand adversity.

Grunblatt E, Mandel S, Youdim MBH. 2000. Neuroprotective Strategies in Parkinson's Disease Using the Models of 6-Hydroxydopamine and MptpVolume 899. p 262-273. Reactive Oxygen Species: From Radiation to Molecular Biology: Annals of the New York Academy of Sciences.
Abstract: The etiology of Parkinson's disease is not known. Nevertheless a significant body of biochemical data from human brain autopsy studies and those from animal models point to an on going process of oxidative stress in the substantia nigra which could initiate dopaminergic neurodegeneration. It is not known whether oxidative stress is a primary or secondary event. Nevertheless, oxidative stress as induced by neurotoxins 6-hydroxydopamine and MPTP (N-methyl-4-phenyl-1,2,3,6-tetrahydropyridine) has been used in animal models to investigate the process of neurodegeneration; with intend to develop antioxidant neuroprotective drugs. It is apparent that in these animal models radical scavengers, iron chelators, dopamine agonists, nitric oxide synthase inhibitors and certain calcium channel antagonists do induce neuroprotection against such toxins if given prior to the insult. Furthermore, recent work from human and animal studies has provided also evidence for an inflammatory process. This expresses itself by proliferation of activated micro glia in the substantia nigra, activation and translocation of transcription factors, NF kappa-beta and elevation of cytotoxic cytokines TNF alpha, IL1-beta, and IL6. Both radical scavengers and iron chelators prevent LPS (lipopolysaccharide) and iron induced activation of NF kappa-B. If an inflammatory response is involved in Parkinson's disease it would be logical to consider antioxidants and the newly developed non-steroid anti-inflammatory drugs such as COX2 (cyclo oxygenase) inhibitors as a form of treatment. However to date there has been little or no success in the clinical treatment of neurodegenerative diseases per se (Parkinson's disease, ischemia etc.), where neurons die, while in animal models the same drugs produce neuroprotection, This may indicate that either the animal models employed are not reflective of the events in neurodegenerative diseases or that because neuronal death involves a cascade of events, a single neuroprotective drug would not be effective. Thus, consideration should be given to multi-neuroprotective drug therapy in Parkinson's disease, similar to the approach taken in AIDS and cancer therapy.

Grunblatt E, Mandel S, Youdim MBH. 2000. MPTP and 6-hydroxydopamine-induced neurodegeneration as models for Parkinson's disease: neuroprotective strategies. J Neurol 247(Suppl. 2):95-102.
Abstract: The etiology of Parkinson's disease is not known. Nevertheless, a significant body of biochemical data from human brain autopsy studies and from animal models points to an ongoing process of oxidative stress in the substantia nigra, which could initiate dopaminergic neurodegeneration. It is not known whether oxidative stress is a primary or secondary event. Oxidative stress, as induced by the neurotoxins 6-hydroxydopamine and MPTP (N-methyl-4-phenyl-1,2,3,6-tetrahydropyridine), has been used in animal models to investigate the process of neurodegeneration to facilitate the development of antioxidant, neuroprotective drugs. It is apparent in these animal models that radical scavengers, iron chelators, dopamine agonists, nitric oxide synthase inhibitors and certain calcium channel antagonists provide neuroprotection against such toxins if given prior to the insult. Furthermore, recent work from human and animal studies has provided evidence of an inflammatory process. This expresses itself as proliferation of activated microglia in the substantia nigra, activation and translocation of transcription factors and neurotrophic factor (NF), kappa-a

and elevation of cytotoxic cytokines, tumour necrosis factor (TNF)-alpha interleukin (IL)-I beta, and IL-6. Both radical scavengers and iron chelators prevent lipopolysaccharide (LPS) and iron-induced activation of NF kappa-beta. If an inflammatory response is involved in Parkinson's disease, it would be logical to consider antioxidants and the newly developed, non-steroidal, anti-inflammatory drugs such as cyclo-oxygenase (COX2) inhibitors as a form of treatment. However, to date there has been little or no success in the clinical treatment of neurodegenerative diseases (for example, Parkinson's disease, ischaemia etc.) where neurons die, while in animal models the same drugs provide neuroprotection. This may indicate that either the animal models employed do not reflect the events in neurodegenerative diseases, or that because neuronal death involves a cascade of events, a single neuroprotective drug is not effective. Thus, consideration should be given to multi-neuroprotective drug therapy in Parkinson's disease, similar to the approach taken in AIDS and cancer therapy.

Graham JM, Paley MNJ, Grunewald RA, Hoggard N, Griffiths PD. 2000. Brain iron deposition in Parkinson's disease imaged using the PRIME magnetic resonance sequence. Brain 123:2423-2431.
Abstract: dIron content of the basal ganglia was investigated in 25 patients with idiopathic Parkinson's disease and 14 matched healthy control subjects using a partially refocused interleaved multiple echo sequence on a 1.5 Tesla MRI system, R-2* (1/T-2*) and R-2' (1/T-2') relaxation rates were higher in the substantia nigra of patients with Parkinson's disease, which indicates that iron content is elevated in this region. R-2' was lower in the putamen, indicating reduced iron levels; reduction in this region was positively correlated with disease duration. Iron-related oxidative stress may contribute to the neurodegeneration of Parkinson's disease, which may lead to alterations in the iron levels of the striatum, We describe a simple, noninvasive technique for measuring iron content.

Gospe SM, Caruso RD, Clegg MS, Keen CL, Pimstone NR, Ducore JM, Gettner SS, Kreutzer RA. 2000. Paraparesis, hypermanganesaemia, and polycythaemia: a novel presentation of cirrhosis. Arch Dis Child 83(5): 439-442.
Abstract: Progressive myelopathy is a rare complication of chronic hepatic disease which has never been reported in the paediatric age group. We describe the 11 year course of an adolescent male with hepatic myelopathy caused by cryptogenic micronodular cirrhosis. His condition has been associated with persistent polycythaemia and extraordinary increases of whole blood manganese, with magnetic resonance imaging evidence of manganese deposition within the basal ganglia and other regions of the brain. The patient has developed neither liver failure nor parkinsonism. The pathophysiological bases of this multiorgan system disorder are described.

Gauthier E, Fortier I, Courchesne F, Pepin P, Mortimer J, Gauvreau D. 2000. Aluminum forms in drinking water and risk of Alzheimer's disease. Environ Res 84(3):232-246.
Abstract: The objective of this study was to assess the relation between long-term exposure to different aluminum (AI) forms in drinking water and Alzheimer's disease (AD). The study participants were selected from a random sample of the elderly population (greater than or equal to 70 years of age) of the Saguenay-lac-Saint-Jean region (Quebec). Sixty-eight cases of Alzheimers disease diagnosed according to recognized criteria were paired for age (+/-2 years) and sex with nondemented controls. Aluminum speciation was assessed using established standard analytical protocols along with quality control procedures. Exposure to Al forms (total Al, total dissolved AI, monomeric organic AI, monomeric inorganic Al, polymeric Al, Al3+, AlOH, AIF, AlH3SiO42-, AlSO4) in drinking water was estimated by juxtaposing the subject's residential history with the physicochemical data of the municipalities. The markers of longterm exposures (1945 to onset) to AZ forms in drinking water were not significantly associated with AD. On the other hand, after adjustment for education level, presence of family

cases, and ApoE epsilon4 allele, exposure to organic monomeric aluminum estimated at the onset of the disease was associated with AD (odds ratio 2.67; 95% CI 1.04-6.90). On average, the exposure estimated at the onset had been stable for 44 years. Our results confirm prime the importance of estimation of Al speciation and consideration of genetic characteristics in the assessment of the association between aluminum exposure and Alzheimer's disease. (C) 2000 Academic Press.

Galvin JE, Giasson B, Hurtig HI, Lee VMY, Trojanowski JQ. 2000. Neurodegeneration with brain iron accumulation, type 1 is characterized by alpha-, beta-, and gamma-synuclein neuropathology. Am J Pathol 157(2): 361-368.
Abstract: Neurodegeneration with brain iron accumulation, type 1 (NBIA 1), or Hallervorden-Spatz syndrome, is a rare neurodegenerative disorder characterized clinically by Parkinsonism, cognitive impairment, pseudobulbar features, as well as cerebellar ataxia, and neuropathologically by neuronal loss, gliosis, and iron deposition in the globus pallidus, red nucleus, and substantia nigra. The hallmark pathological lesions of NBIA 1 are axonal spheroids, but Lewy body (LB)-like intraneuronal inclusions, glial inclusions, and rare neurofibrillary tangles also occur. Here we show that there is an accumulation of alpha-synuclein (alpha S) in LB-like inclusions, glial inclusions, and spheroids in the brains of three NBIA 1 patients. Further, beta-synuclein (beta S) and gamma-synuclein (gamma S) immunoreactivity was detected in spheroids but not in LB-like or glial inclusions. Western blot analysis demonstrated high-molecular weight alpha S aggregates in the high-salt-soluble and Triton X-100-insoluble/sodium dodecyl sulfate-soluble fraction of the NBIA 1 brain. Significantly, the levels of alpha S were markedly reduced in the Triton X-100-soluble fractions compared to control brain, and unlike other synucleinopathies, insoluble alpha S did not accumulate in the formic acid-soluble fraction. These findings expand the concept of neurodegenerative synucleinopathies by Implicating alpha S, beta S, and gamma S in the pathogenesis of NBIA 1.

Fredriksson A, Schroder N, Eriksson P, Izquierdo I, Archer T. 2000. Maze learning and motor activity deficits in adult mice induced by iron exposure during a critical postnatal period. Developmental Brain Research 119(1):65-74.
Abstract: Newborn mice were administered Fe2+ (iron succinate: 7.5 mg/kg, b.wt) on either Days 3-5, 10-12 or 19-21, or vehicle (saline) at the same times, postnatally. Spontaneous motor behaviour and radial arm maze learning were tested at the age of 3 months. It was found that mice treated with Fe2+ during postnatal Days 10-12 were markedly hypokinetic during the 1st 20-min test period and hyperkinetic during the 3rd and final 20-min test period. These mice showed an almost complete lack of habituation of spontaneous motor activity parameters to the test chambers. In the radial arm maze, the Days 10-12 treatment group evidenced significantly both more errors in arm choices and longer latencies to acquire all eight pellets; these mice showed also a severe trial-to-trial retention deficit as indexed by retention quotients. These behavioural deficits were observed also in animals treated with Fe2+ during postnatal Days 3-5, but the effects were less pronounced, indicating the higher susceptibility of the brain for Fe2+-induced damage during Days 10-12 postpartum. Treatment with Fe2+ on Days 19-21 did not induce behavioural alterations in comparison with its respective control (vehicle) group. Analysis of total brain iron content indicated significantly more iron (mu g/g) accumulation in the basal ganglia, but not frontal cortex, of mice from the Days 3-5 and 10-12 Fe2+ (7.5 mg/kg) treatment groups. The contribution of iron overload during the immediate postnatal to later functional deficits seems to implicate symptoms of Parkinsonism but the kinetics of iron uptake to the brain and its regional distribution at this critical period of development awaits elucidation. (C) 2000 Elsevier Science B.V. All rights reserved.

Frankel D, Mehindate K, Schipper HM. 2000. Role of heme oxygenase-1 in the regulation of manganese superoxide dismutase gene expression in

oxidatively-challenged astroglia. J Cell Physiol 185(1):80-86.
Abstract: Manganese superoxide dismutase (MnSOD) is an antioxidant enzyme that reduces superoxide anion to hydrogen peroxide in cell mitochondria. MnSOD is overexpressed in normal aging brain and in various central nervous system disorders; however, the mechanisms mediating the upregulation of MnSOD under these conditions remain poorly understood. We previously reported that cysteamine (CSH) and other pro-oxidants rapidly induce the heme oxygenase-1 (HO-1) gene in cultured rat astroglia followed by late upregulation of MnSOD in these cells. In the present study, we demonstrate that antecedent upregulation of HO-1 is necessary and sufficient for subsequent induction of the MnSOD gene in neonatal rat astroglia challenged with CSH or dopamine, and in astroglial cultures transiently transfected with full-length human HO-1 cDNA. Treatment with potent antioxidants attenuates MnSOD expression in HO-1-transfected astroglia, strongly suggesting that intracellular oxidative stress signals MnSOD gene induction in these cells. Activation of this HO-1-MnSOD axis may play an important role in the pathogenesis of Alzheimer disease, Parkinson disease and other free radical-related neurodegenerative disorders. In these conditions, compensatory upregulation of MnSOD may protect mitochondria from oxidative damage accruing from heme-derived free iron and carbon monoxide liberated by the activity of HO-1. (C) 2000 Wiley-Liss, Inc.

Foley P, Riederer P. 2000. Influence of neurotoxins and oxidative stress on the onset and progression of Parkinson's disease. J Neurol 247(Suppl 2):82-94.
Abstract: It is generally accepted that progressive, irreversible and regionally specific neurodegeneration and the presence of Lewy bodies are the essential pathological hallmarks of idiopathic parkinsonism. The causes of these phenomena, however, remain to be elucidated. One of the leading hypotheses is that oxidative stress induced by reactive oxygen species (ROS), such as the hydroxyl radical, damages essential components of the neuron, resulting ultimately in cell death. Observations in the parkinsonian brain at post-mortem support this hypothesis; for example, widespread oxidative protein modification is evident. There are several potential sources of increased oxidative stress in Parkinson's disease, including mitochondrial dysfunction, increased foe iron levels and impaired free radical defensive mechanisms. Further, it is possible that glial, rather than neuronal, elements are primarily responsible for the initial increase in oxidative stress in the substantia nigra. It is likely that parkinsonism is the result of aberrations at multiple levels of neuronal function. Oxidative stress is no doubt one of the events involved in neurodegeneration, but is unlikely to be the initiating event. It is to be expected that the search for this event will continue for many years.

Floris G, Medda R, Padiglia A, Musci G. 2000. The physiopathological significance of ceruloplasmin - A possible therapeutic approach. Biochem Pharmacol 60 (12):1735-1741.
Abstract: This article reviews and comments on the physiological roles of ceruloplasmin (Cp). We show that, in addition to its ascertained involvement in iron homeostasis, the protein, by virtue of its unique structure among multicopper oxidases, is likely involved in other processes of both an enzymatic and a nonenzymatic nature. In particular, based on the analysis of the kinetic parameters, on the one hand, and of the side-products of the oxidation, on the other, we propose that the long-recognized ability of Cp to interact with and oxidize non-iron substrates may be of physiological relevance. The striking example of 6-hydroxydopamine oxidation is presented, where we show that the catalytic action is carried out readily under physiological conditions, without release of potentially toxic oxygen intermediates. BIOCHEM PHARMACOL 60;12:1735-1741, 2000. (C) 2000 Elsevier Science Inc.

Floor E. 2000. Iron as a vulnerability factor in nigrostriatal degeneration in aging and Parkinson's disease. Cell Mol Biol 46(4):709-720.
Abstract: Recent findings strengthen the connection between iron accumulation in the basal ganglia, oxidative stress and nigrostriatal

degeneration. Oxidative stress appears to be elevated in the normal human substantia nigra in comparison with other brain regions, and further increases occur in Parkinson's disease. Accumulation of iron may contribute to degeneration of nigral dopamine neurons by catalyzing oxidative damage to cell components and also by perturbing the network of interactions that modulate cellular redox status.

Fernandez-Gonzalez A, Perez-Otano I, Morgan JI. 2000. MPTP selectively induces haem oxygenase-1 expression in striatal astrocytes. Eur J Neurosci 12(5): 1573-1583.
Abstract: Parkinson's disease (PD) is characterized by the loss of dopaminergic neurons in the substantia nigra pars compacta with accompanying evidence of increased oxidative damage, deficits in mitochondrial function and iron deposition. Recently, haem oxygenase-1 levels were reported to be elevated in PD brains. Because this enzyme is involved in the response to oxidative stress and is critical for cellular haem and iron homeostasis, it could play a role in the pathogenesis of PD. Therefore, we investigated the expression of haem oxygenase isoform 1 (HO-1) in the 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) mouse model of PD. MPTP triggered a relatively rapid and persistent increase in HO-1 mRNA exclusively in the mouse striatum. In situ hybridization and immunohistochemistry showed HO-1 to be localized to striatal astrocytes. The induction of HO-1 by MPTP was blocked by selegiline and GBR-12909, indicating the protoxin had to be metabolized by monoamine oxidase B and taken up by dopaminergic neurons to exert its action in astrocytes. MPTP did not alter the expression of other enzymes of haem synthesis or degradation nor were the levels of mRNA for haem or iron-binding proteins changed. Thus, expression of HO-1 was not part of a cellular program involving haem biosynthesis or homeostasis. In addition, heat shock proteins were not induced by MPTP. Thus, MPTP elicited a selective transcriptional response in striatal astrocytes. This response appears to be mediated by molecules released from affected dopaminergic nerve terminals in the striatum acting upon neighbouring astrocytes. This signalling pathway and its potential relevance to PD are discussed.

Feany MB. 2000. Studying human neurodegenerative diseases in flies and worms. J Neuropathol Exp Neurol 59(10):847-856.
Abstract: Invertebrate models of several human neurodegenerative diseases have recently been described. These models faithfully replicate key neuropathological features of the human disorders. Because the basic cell biology of the nervous system is very similar in vertebrates and invertebrates, the sophisticated and rapid genetic analysis feasible in Drosophila and C. elegans promises significant insight into human neurodegenerative syndromes. In addition, the short lifespan, small size, and ease of culturing make worms and flies ideal for drug testing.

Ebadi M, Rojas P, Hiramatsu M, Pellett L, Muralikrishnan D, Murphy T, Drees K. 2000. Metallothionein(MT) isoforms protect against MPTP-induced parkinsonism. J Neurochem 74:S16.

Duda JE, Lee VMY, Trojanowski JQ. 2000. Neuropathology of synuclein aggregates: New insights into mechanisms of neurodegenerative diseases. J Neurosci Res 61(2):121-127.
Abstract: Beginning with the isolation of the fragment of alpha-synuclein (alpha-syn) known as the non-A beta component of amyloid plaques (NAC peptide) from Alzheimer's disease (AD) brains, alpha-syn has been increasingly implicated in the pathogenesis of neurodegenerative diseases, which now are classified as synucleinopathies. Indeed, unequivocal evidence linking abnormal alpha-syn to mechanisms of brain degeneration came from discoveries of missense mutations in the alpha-syn gene pathogenic for familial Parkinson's disease (PD) in rare kindreds. Shortly thereafter, alpha-syn was shown to be a major component of Lewy bodies (LBs) and Lewy neurites in sporadic PD, dementia with LBs (DLB) and the LB variant of AD. Also, studies of brains from patients with AD caused by genetic abnormalities demonstrated many a-syn positive LBs, Further,

alpha-syn was implicated in the formation of the glial (GCIs) and neuronal cytoplasmic inclusions of multiple system atrophy, and the LBs, GCIs and neuraxonal spheroids of neurodegeneration with brain iron accumulation type I. Recently, two other members of the synuclein family, beta- and gamma-synuclein, have also been recognized to play a role in the pathogenesis of novel axonal lesions in PD and DLB. Evidence for a role of a-syn in the formation of filamentous aggregates was reinforced by in vitro studies showing aggregation and fibrillogenesis of mutant and wild type alpha-syn. Indeed, since the aggregation of brain proteins into presumptively toxic lesions is emerging as a common but poorly understood mechanistic theme in sporadic and hereditary neurodegenerative diseases, clarification of the mechanism of synuclein aggregation could augment efforts to develop novel and more effective therapies for many neurodegenerative disorders. (C) 2000 Wiley-Liss, Inc.

Duda JE, Giasson BI, Chen QP, Gur TL, Hurtig HI, Stern MB, Gollomp SM, Ischiropoulos H, Lee VMY, Trojanowski JQ. 2000. Widespread nitration of pathological inclusions in neurodegenerative synucleinopathies. Am J Pathol 157(5):1439-1445.
Abstract: Reactive nitrogen species may play a mechanistic rot in neurodegenerative diseases by posttranslationally altering normal brain proteins. In support of this hypothesis, we demonstrate that an anti-3-nitrotyrosine polyclonal antibody stains all of the major hallmark lesions of synucleinopathies Including Lewy bodies, Lewy neurites and neuraxonal spheroids in dementia with Lewy bodies, the Lewy body variant of Alzheimer's disease, and neurodegeneration with brain iron accumulation type 1, as well. as glial and neuronal cytoplasmic inclusions in multiple system atrophy. This antibody predominantly recognized nitrated alpha -synuclein when compared to other In vitro nitrated constituents of these pathological lesions, such as neurofilament subunits and microtubules, Collectively, these findings imply that alpha -synuclein is nitrated in pathological lesions. The widespread presence of nitrated alpha -synuclein in diverse intracellular inclusions suggests that oxidation/nitration is involved in the onset and/or progression of neurodegenerative diseases.

Double KL, Gerlach M, Youdim MB, Riederer P. 2000. Impaired iron homeostasis in Parkinson's disease. J Neural Transm Suppl (60):37-58.
Abstract: Despite physiological systems designed to achieve iron homeostasis, increased concentrations of brain iron have been demonstrated in a range of neurodegenerative diseases. These including the parkinsonian syndromes, the trinucleotide repeat disorders and the dementia syndromes. The increased brain iron is confined to those brain regions most affected by the degeneration characteristic of the particular disorder and is suggested to stimulate cell damage via oxidative mechanisms. Changes in central iron homeostasis have been most closely investigated in PD, as this disorder is well characterised both clinically and pathologically. PD is associated with a significant increase in iron in the degenerating substantia nigra (SN) and is measureable in living PD patients and in post-mortem brain. This increase, however, occurs only in the advanced stages of the disease, suggesting that this phenonoma may be a secondary, rather than a primary initiating event, a hypothesis also supported by evidence from animal experiments. The source of the increased iron is unknown but a variety of changes in iron homeostasis have been identified in PD, both in the brain and in the periphery. The possibility that an increased amount of iron may be transported into the SN is supported by data demonstrating that one form of the iron-binding glycoprotein transferrin family, lactotransferrin, is increased in surviving neurons in the SN in the PD brain and that this change is associated with increased numbers of lactotransferrin receptors on neurons and microvessels in the parkinsonian SN. These changes could represent one mechanism by which iron might concentrate within the PD SN. Alternatively, the measured increased in iron might result from a redistribution of ferritin iron stores. Ferritin is located in glial cells while the degenerating neurons do not stain positive for ferritin. As free radicals are highly reactive, it is unlikely that glial-derived free radicals diffuse across

the intracellular space in sufficent quantities to damage neuronal constituents. If intracellular iron release contributes to neuronal damage it seems more probable that an intraneuronal iron source is responsible for oxidant-mediated damage. Such a iron source is neuromelanin (NM), a dark-coloured pigment found in the dopaminergic neurons of the human SN. In the normal brain, NM has the ability to bind a variety of metals, including iron, and increased NM-bound iron is reported in the parkinsonian SN. The consequences of these phenomena for the cell have not yet been clarified. In the absence of significant quantities of iron NM can act as an antioxidant, in that it can interact with and inactivate free radicals. On the other hand, in the presence of iron NM appears to act as a proxidant, increasing the rate of free radical production and thus the oxidative load within the vulnerable neurons. Given that increased iron is only apparent in the advanced stages of the disease it is unlikely that NM is of importance for the primary aetiology of PD. A localised increase in tissue iron and its interaction with NM may be, however, important as a secondary mechanism by increasing the oxidative load on the cell, thereby driving neurodegeneration.

Dorman DC. 2000. An integrative approach to neurotoxicology. Toxicol Pathol 28 (1):37-42.
Abstract: Exposure of human populations to a wide variety of chemicals has generated concern about the potential neurotoxicity of new and existing chemicals. Experimental studies conducted in laboratory animals remain critical to the study of neurotoxicity. An integrative approach using pharmacokinetic, neuropathological, neurochemical, electrophysiological, and behavioral methods is needed to determine whether a chemical is neurotoxic. There are a number of factors that can affect the outcome of a neurotoxicity study, including the choice of animal species, dose and dosage regimen, route of administration, and the intrinsic sensitivity of the nervous system to the test chemical. The neurotoxicity of a chemical can vary at different stages of brain development and maturity. Evidence of neurotoxicity may be highly subjective and species specific and can be complicated by the presence of systemic disease. The aim of this paper is to give an overview of these and other factors involved in the assessment of the neurotoxic potential for chemicals. This article discusses the neurotoxicity of several neurotoxicants (eg, acrylamide, trimethyltin, 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine, manganese, and ivermectin), thereby highlighting a multidisciplinary approach to the assessment of chemically induced neurotoxicity in animals. These model chemicals produce a broad range of effects that includes peripheral axonopathy, selective neuronal damage within the nervous system, and impaired neuronal-glial metabolism.

Discalzi G, Pira E, Hernandez EH, Valentini C, Turbiglio M, Meliga F. 2000. Occupational Mn parkinsonism: Magnetic resonance imaging and clinical patterns following CaNa2 -EDTA chelation. Neurotoxicology 21(5):863-866.
Abstract: We report a case of occupational parkinsonism due to manganese exposure in which professional exposure has been documented both by the high blood and urinary levels of the metal and by its presence in the materials used. A strong relation was evident among chelating treatment, cessation of exposure and clinical improvement. MRI confirmed the! evolution of clinical pattern by means of reduction of heavy metal deposition in basal ganglia. These findings also agree with the few experimental and human studies published. This case study points to the need for an accurate occupational history collection and suggests the possibility of useful chelating therapy with CaNa(2)EDTA. (C) 2000 Inter Press, Inc.

Dietz MC, Wrazidlo W, Ihrig A, Bader M, Triebig G. 2000. Magnetic resonance imaging (MRI) of the central nervous system in long-term manganese dioxide (MnO2) exposed workers. Rofo-Fortschritte Auf Dem Gebiet Der Rontgenstrahlen Und Der Bildgebenden Verfahren 172(6):514-520.
Abstract: Aim: Changes within the brain detected by MRI after chronic manganese poisoning raised the question whether morphological changes

of the basal ganglia, particularly of the globus pallidus, could be detected after chronic occupational exposure to manganese dioxide. Method: In a cross-sectional study, healthy workers (48 male and 27 female) at a dry cell battery factory were examined. Actual internal exposure was quantified by the analysis of manganese in the blood using atomic absorption spectrometry. Chronic exposure was defined as a cumulative index (CBI) including duration of exposure, individual workplace factors, and previously measured concentrations of MnO2 in dust samples. A Philips Gyroscan T5-II (0.5 T) was used for the MRI of the brain. The following indicators were taken to ascertain possible manganese-induced changes; Pallidum-Index (PI), width of 3(rd) ventricle and cella media index in addition to clinical examinations. Results: No cases of parkinsonism were detected in clinical examinations or by other means. The mean manganese concentration in blood was 12 mu g/l (range: 3.9-23.3 mu g/l). In comparison to the upper reference value of 10 mu g/l, 42 workers (56%) had a higher body burden. A significant positive correlation between manganese levels in blood and the PI (indicated by T-1-shortening) was observed as well as between the CBI and workplace-specific exposure. Brain atrophy was not detected in any of the observed cases. Conclusions: Long-term exposure to manganese dioxide dust correlates with the Pallidum-Index in MRI scans. Although the MRI findings have no current clinical relevance for individuals, further studies are necessary to evaluate specificity and potential prognostic value.

Demougeot C, Marie C, Beley A. 2000. Importance of iron location in iron-induced hydroxyl radical production by brain slices. Life Sci 67(4):399-410.
Abstract: Iron imbalance has been implicated in oxidative injury associated with many brain diseases. The present study investigated the importance of iron location in hydroxyl radical ((OH)-O-.) generation and the link between (OH)-O-. production evaluated by the salicylate method and lipid peroxidation monitored by thiobarbituric acid-reactive substances assay. Brain slices were exposed to increasing doses (2, 10 and 50 mu M) of Fe (III) that was complexed either to a lipophilic (8-hydroxyquinoline, HQ) or to a hydrophilic (ammoniacal citrate) ligand, Both iron complexes resulted in an increased salicylate hydroxylation and lipid peroxidation, these effects being significantly more potent in presence of Fe(III)-HQ. Salicylate hydroxylation was linearly correlated to the intensity of TEARS formation but the slope of the curve was found to be higher with Fe(III)-HQ. The present results demonstrate that 1) cell-associated reactive iron is more pr one than extracellular iron to induce (OH)-O-. generation, 2) the level of lipid peroxidation depending on the site of (OH)-O-. production. cannot be used as an index of the level of total (OH)-O-. formation, 3) the salicylate method is a convenient method to detect (OH)-O-. formed intracellularly, at least in vitro. (C) 2000 Elsevier Science Inc. All rights reserved.

Demarquay G, Setiey A, Morel Y, Trepo C, Chazot G, Broussolle E. 2000. Clinical report of three patients with hereditary hemochromatosis and movement disorders. Mov Disord 15(6):1204-1209.
Abstract: Neurologic manifestations are rarely described in hereditary hemochromatosis (HH). We describe three patients with HH and movement disorders. Patient 1, a 69-year-old man, had a 13-year history of disabling cerebellar syndrome, action tremor and myoclonus, and secondary dementia. Patient 2 was a 40-year-old man with a 9-year history of cerebellar syndrome, head and arm tremor, and cervical dystonia. Patient 3, a 75-year-old woman, had a 5-year history of rapidly disabling parkinsonian syndrome unresponsive to levodopa. The diagnosis of HH was established in the three patients by iron tests, evidence of a C282Y mutation, and, in two patients, by liver biopsy. High-field T2-weighted magnetic resonance imaging showed hyperintense signals in hemispheric white matter in patient 1, cerebellar atrophy in patient 2, and cerebellar and cerebral atrophy in patient 3 and no significant hypointense signals in the three patients. Phlebotomies and symptomatic treatments did not change the course of the disease. Our cases are compared with the five previously reported observations of HH with movement disorders. This rare association is one cause of the chronic acquired non-Wilsonian

hepatocerebral degeneration syndromes and represents a separate entity from aceruloplasminemia. The pathophysiologic mechanism of movement disorders in HH is unresolved. No hepatic insufficiency and portosystemic encephalopathy is evidenced in our cases, whereas the putative role of abnormal iron load remains to be ascertained. HH should be investigated more systematically in patients with movement disorders.

Delisle MB, Uro-Coste E, Murrell JR, Rascol O, Ghetti B. 2000. Neurodegenerative disease with a mutation at codon 279 (N279 K) in exon 10 of the protein Tau gene. Bull Acad Natl Med 184(4):799-809; discussion 809-811.
Abstract: Frontotemporal dementia and Parkinsonism linked to chromosome 17 (FTDP-17) are related to pathogenic mutations of the Tau gene. One of these, located at codon 279, results in an asparagine to lysine substitution. It was detected in three unrelated families from different origins. This maturation affects ater splicing, allowing exon 10 to be incorporated more frequently in the Tau transcripts, causing an abnormal preponderance of three-over four-repeat isoforms in soluble tau and the presence of the four-repeat isoforms in the insoluble tau. To better understand this newly described pathology, we analysed data from the three previously reported families. The American family, described as "pallido-ponto-nigral degeneration" is a large family which has been extensively studied (13 neuropathological studies). The Japanese family was initially presented as "pallidonigroluysuan degeneration with iron deposition" and recently found to be related to N279 K mutation. We reported clinical, pathological and genetic data from the French family. Clinical particularities are ocular movements alterations with vertical supranuclear palsy, extrapyramidal signs (rigidity, dyskinesia, with atypical resting and postural tremor) and progressive dementia. Partial or no L-DOPA responsiveness is noted. These features led to discuss progressive supranuclear palsy, in some cases. There is no amyotrophy, nor any sensibility to neuroleptics, both signs being observed on other FTDP-17 syndromes. Neuropathology and immunohistochemistry confirm the presence of Tau immunolabeled inclusions, affecting mainly neurons in grain stem nuclei and glial cells in supratentorial white matter. neuronal loss, which is moderate in frontal and temporal cortex, is severe in substantia nigra and globus pallidum. It is variable in other subcortical structures. In these structures, it is associated with iron deposition. This latter may participate in the degenerative process of cells and led to death in some specific neurons. The selectivity of neuronal death in hereditary diseases, when compared to data concerning sporadic neurodegenerative diseases which share similar clinical signs and neuropathological lesions, reinforces the hypothesis of an increased vulnerability of some neuronal populations which express specific sets tau isoforms. Neurons particularly involved in these diseases express exclusively exon 10 + tau isoforms.

Del Rio MJ, Velez-Pardo C. 2000. Molecular mechanism of monoamine toxicity in Parkinson's disease: hypothetical cell death model. Med Hypotheses 54(2): 269-274.
Abstract: Although there have been experimental approaches to understanding the etiology of Parkinson's disease, the cause of cell degeneration in this neurological disorder remains a mystery. Herein, a hypothetical model is proposed to explain the mechanism leading neurons to die. The model is based on recent experimental evidence and it attempts to dissect the actions of dopamine and metal ions as potential triggers for the activation of an ordered cascade of events of the cell death machinery. (C) 2000 Harcourt Publishers Ltd.

Del Rio MJ, Velez-Pardo C. 2000. 17 beta-Estradiol protects lymphocytes against dopamine and iron-induced apoptosis by a genomic-independent mechanism Implication in Parkinson's disease. General Pharmacology-the Vascular System 35(1):1-9.
Abstract: Dopamine (DA) in combination with iron (Fe2+) has been demonstrated to induce apoptosis in neuronal-like PC12 cells by an oxidative stress mechanism. To get a better insight of cell death and protective mechanisms in DA/Fe2+-induced toxicity, we investigated the

effects of DA/Fe2+ and the antioxidant action of 17 beta -estradiol (E2) in peripheral blood lymphocytes (PBL). We found that DA/Fe2+-induces apoptosis in PBL via a hydrogen peroxide (H2O2)-mediated oxidative mechanism, which in turn triggers a cascade of molecular events requiring RNA and de novo protein synthesis. We have also demonstrated that E2 prevents significantly DA/Fe2+-induced apoptosis in PBL by directly inhibiting the intracellular accumulation of peroxides generated by DA/Fe2 +-reaction. This protective activity is independent of the presence or activation of the estrogen receptors (ERs). These data further support and validate our previous hypothesis that DA/Fe2+/H2O2 could be a general mediator of oxidative stress through a common cell death mechanism in both neuronal and nonneuronal cells. These findings may be particularly relevant to the potential approaches to rescue and prolong the survival of neurons by estrogens in patients with Parkinson's disease (PD). (C) 2001 Elsevier Science Inc. All rights reserved.

Degner D, Bleich S, Riegel A, Sprung R, Poser W, Ruther E. 2000. A follow-up study in enteral manganese intoxication: clinical, laboratory, and neuroradiological aspects. Nervenarzt 71(5):416-419.
Abstract: Manganese intoxication is an unusual, severe form of intoxication. This report deals with a patient now 80 years old who accidentally ingested a solution of potassium permanganate for a period of at least 4 weeks 14 years ago. Since then, the patient suffers from a mild parkinsonian syndrome and distally accentuated polyneuropathies. Psychiatric disorders, especially demential or depressive symptoms, were not observed. Manganese analysis of his hair still shows a clear increase in manganese concentration. The MRI of his brain showed no pathological changes, in particular none of those often described with symmetric signal elevation in T-1 in the area of the basal ganglia. In this study, we present clinical, laboratory, and neuroradiological findings. Unusual in this case with a short exposition is the long duration and clinical improvement without I-dopa treatment.

Copin JC, Gasche Y, Chan PH. 2000. Overexpression of copper/zinc superoxide dismutase does not prevent neonatal lethality in mutant mice that lack manganese superoxide dismutase. Free Radic Biol Med 28(10):1571-1576.
Abstract: There are two types of intracellular superoxide dismutases: the mitochondrial manganese SOD (MnSOD) and the cytoplasmic copper/zinc SOD (CuZnSOD). Mutant mice that lack MnSOD die shortly after birth because of cardiomyopathy and mitochondrial injury. In order to verify if CuZnSOD could compensate for MnSOD deficiency, a new mutant mouse that overexpresses CuZnSOD but is deficient in MnSOD was generated by crossing MnSOD knockout mice with CuZnSOD transgenic mice. CuZnSOD activity was significantly increased in the blood brain, liver, and heart of MnSOD knockout, CuZnSOD transgenic mice when compared with nontransgenic mice. However, overexpression of CuZnSOD did not prevent neonatal lethality in mice that lack MnSOD, nor did it prevent oxidative aconitase inactivation, nor did it rescue MnSOD-deficient astrocytes in culture. Based on our findings, which emphasize the strong enzymatic compartmentalization of CuZnSOD and MnSOD, therapeutic antioxidant strategies should consider the final intracellular localization of the antioxidant used, especially when those strategies are directed against mitochondrial diseases. (C) 2000 Elsevier Science Inc.

Chung JM, Chang SY, Kim YI, Shin HC. 2000. Zinc increases the excitability of dopaminergic neurons in rat substantia nigra. Neurosci Lett 286(3): 183-186.
Abstract: The effect of zinc ions (Zn2+) on the neuronal excitability of substantia nigra (SN) where the zinc level is known higher in Parkinsonian brains than that in normal brains has not yet been elucidated. We, therefore, examined the effect of Zn2+ on the intrinsic electrical properties of dopaminergic SN neurons, using a whole-cell recording method. Zn2+ hyperpolarized dopaminergic SN neurons at resting state. Also Zn2+ shortened the duration of evoked spikes, developed a fast afterhyperpolarization, and increased their firing frequency. Voltage-clamp

studies showed that Zn2+ decreased 4-aminopyridine-sensitive outward currents, suggesting that a transient A-like potassium channel be one of the major targets Zn2+ can modulate in the SN neurons. (C) 2000 Published by Elsevier Science Ireland Ltd. All rights reserved.

Chen MT, Sheu JY, Lin TH. 2000. Protective effects of manganese against lipid peroxidation. Journal of Toxicology and Environmental Health-Part a 61(7): 569-577.
Abstract: The aim of this study was to investigate the effects of chronic, daily, 30-d administration of manganese chloride (MnCl2) to male Sprague-Dawley rats on lipid peroxidation in various tissues. Rats were intraperitoneally injected with MnCl2 (20 mg/kg) once daily for 30 consecutive days. The Mn accumulated in liver, spleen, adrenal glands, heart, kidneys, lung, and testes. This was associated with decreased lipid peroxidation in liver, spleen, and adrenal glands and a decrease in the levels of Fe in these tissues. In a second group of animals, Mn (20 mg/kg/d) and glutathione (GSH, 15 mg/kg/d) were administered ip for 30 d. GSH counteracted the Mn-induced protective fall in lipid peroxidation, but Fe levels remained lower in liver and spleen. Mn decreases lipid peroxidation in certain tissues, which may involve lowering Fe content, but interaction with Fe is not the sole mechanism.

Chaki H, Furuta S, Matsuda A, Yamauchi K, Yamamoto K, Kokuba Y, Fujibayashi Y. 2000. Magnetic resonance image and blood manganese concentration as indices for manganese content in the brain of rats. Biol Trace Elem Res 74 (3):245-257.
Abstract: Neurological disorders similar to parkinsonian syndrome and signal hyperintensity in brain on TI-weighted magnetic resonance (MR) images have been reported in patients receiving long-term total parenteral nutrition (TPN). These symptoms have been associated with manganese (Mn) depositions in brain. Although alterations of signal intensity on T-1-weighted MR images in brain and of Mn concentration in blood are theoretically considered good indices for estimating Mn deposition in brain, precise correlations between these parameters have not been demonstrated as yet. Male Sprague-Dawley rats received TPN with 10-fold the clinical dose of the trace element preparation (TE-5) for 7 d. At 0, 2, 4, 6, and 8 wk post-TPN, the cortex, striatum, midbrain, and cerebellum were evaluated by MR images, and Mn concentration in blood and Mn content in these brain sites were measured by atomic absorption spectrometry. Immediately after TPN termination, signal hyperintensity in brain sites and elevated Mn content in blood and brain sites were observed. These values recovered at 4 wk post-TPN. A positive correlation was observed between either the signal intensity in certain brain sites or Mn content in blood and the relevant brain sites.

Castellani RJ, Siedlak SL, Perry G, Smith MA . 2000. Sequestration of iron by Lewy bodies in Parkinson's disease. Acta Neuropathol (Berl) 100(2): 111-114.
Abstract: Central to the oxidative stress hypothesis of Parkinson's disease (PD) pathogenesis is the ability of iron to generate hydroxyl radicals via the Fenton reaction, and the consistent demonstration of iron elevation in the pars compacta region of the substantia nigra. However, uncertainty exists as to whether the excess iron exists in a state suitable for redox chemistry. Here, using a method we developed that detects redox-active iron in situ, we were able to demonstrate strong labeling of Lewy bodies in substantia nigra pars compacta neurons in PD. In contrast, cortical Lewy bodies in cases of Lewy body variant of Alzheimer's disease were unstained. While the presence of elevated iron in PD substantiates the oxidative stress hypothesis, one must remember that these are viable neurons, indicating that Lewy bodies may act to sequester iron in PD brains in a protective, rather than degenerative, mechanism. The absence of redox-active iron in neocortical Lewy bodies highlights a fundamental difference between cortical and brain stem Lewy bodies.

Camandola S, Poli G, Mattson MP. 2000. The lipid peroxidation product 4-

hydroxy-2,3-nonenal inhibits constitutive and inducible activity of nuclear factor kappa B in neurons. Molecular Brain Research 85(1-2):53-60.
Abstract: Peroxidation of membrane lipids occurs in many different neurodegenerative conditions including stroke, and Alzheimer's and Parkinson's diseases. Recent findings suggest that lipid peroxidation can promote neuronal death by a mechanism involving production of the toxic aldehyde 4-hydroxy-2,3-nonenal (HNE), which may act by covalently modifying proteins and impairing their function. The transcription factor NF-kappaB can prevent neuronal death in experimental models of neurodegenerative disorders by inducing the expression of anti-apoptotic proteins including Bcl-2 and manganese superoxide dismutase. We now report that HNE selectively suppresses basal and inducible NF-kappaB DNA binding activity in cultured rat cortical neurons. Immunoprecipitation-immunoblot analyses using antibodies against HNE-conjugated proteins and p50 and p65 NF-kappaB subunits indicate that HNE does not directly modify NF-kappaB proteins. Moreover, HNE did not affect NF-kappaB DNA-binding activity when added directly to cytosolic extracts, suggesting that HNE inhibits an upstream component of the NF-kappaB signaling pathway. Inhibition of the survival-promoting NF-kappaB signaling pathway by HNE may contribute to neuronal death under conditions in which membrane lipid peroxidation occurs. (C) 2000 Elsevier Science BN. All rights reserved.

Calingasan NY, Gibson GE. 2000. Dietary restriction attenuates the neuronal loss, induction of heme oxygenase-1 and blood-brain barrier breakdown induced by impaired oxidative metabolism. Brain Res 885(1):62-69.
Abstract: Experimental thiamine deficiency (TD) is a model of impaired oxidative metabolism associated with region-selective neuronal loss in the brain. Oxidative stress is a prominent feature of TD neuropathology, as evidenced by the accumulation of heme oxygenase-1 (HO-1), ferritin, reactive iron and superoxide dismutase in microglia, nitrotyrosine and 4-hydroxynonenal in neurons, as well as induction of endothelial nitric oxide synthase within the vulnerable areas. Dietary restriction (DR) reduces oxidative stress in several organ systems including the brain. DR increases lifespan and reduces neurodegeneration in a variety of models of neuronal injury. The possibility that DR can protect vulnerable neurons against TD-induced oxidative insults has not been tested. The current studies tested whether approximately 3 months of DR (60% of ad libitum intake) altered the response to TD. Six month-old ad libitum-fed or dietary restricted C57BL/6 mice received a thiamine-deficient diet either ad libitum, or under a DR regimen respectively for eleven days. The TD mice also received daily injections of the thiamine antagonist pyrithiamine. Control ad libitum-fed or DR mice received an unlimited amount, or 60% of ad libitum intake, respectively, of thiamine-supplemented diet. As in past studies, TD produced region-selective neuronal loss (-60%), HO-1 induction, and IgG extravasation in the thalamus of ad libitum-fed mice. DR attenuated the TD-induced neuronal loss (-30%), HO-1 induction and IgG extravasation in the thalamus. These studies suggest that oxidative damage is critical to the pathogenesis of TD, and that DR modulates the extent of free radical damage in the brain. Thus, TD is an important model for studying the relationship between aging, oxidative stress and nutrition. (C) 2000 Elsevier Science B.V. All rights reserved.

Bush AI. 2000. Metals and neuroscience. Curr Opin Chem Biol 4(2):184-191.
Abstract: Data are now rapidly accumulating to show that metallochemical reactions might be the common denominator underlying Alzheimer's disease, amyotrophic lateral sclerosis, prion diseases, cataracts, mitochondrial disorders and Parkinson's disease. In these disorders, an abnormal reaction between a protein and a redox-active metal ion (copper or iron) promotes the formation of reactive oxygen species or radicalization. It is especially intriguing how the powerful catalytic redox activity of antioxidant Cu/Zn-superoxide dismutase can convert into a pro-oxidant activity, a theme echoed in the recent proposal that A beta and PrP, the proteins respectively involved in Alzheimer's disease and prion diseases, possess similar redox activities.

Bi SP, Zhang FP, Zou GW. 2000. Progresses of studies of aluminum intoxication in the pathology of Alzheimer disease, Parkinson disease, dialysis encephalopathy, bone disease and anaemia. Abstracts of Papers of the American Chemical Society 220:U189.

Berkovitch M, Bistritzer T, Milone SD, Perlman K, Kucharczyk W, Koren G, Olivieri NF. 2000. Iron deposition in the anterior pituitary in homozygous beta-thalassemia: MRI evaluation and correlation with gonadal function. Journal of Pediatric Endocrinology & Metabolism 13(2):179-184.
Abstract: Objective: Iron deposition in the anterior pituitary continues to pose a serious problem in older patients with homozygous beta-thalassemia particularly in terms of gonadal function. This study aimed to investigate whether iron loading within the pituitary correlated with endocrine function. Patients: 33 patients above 15 years of age, with transfusion-dependent homozygous beta-thalassemia and iron overload were studied. All had been receiving deferoxamine since 1978, Design and Measurements: The endocrine status of the patients was assessed on clinical examination by an endocrinologist, and by a gonadotropin releasing hormone stimulation test. MRI of the pituitary was carried out for each patient. Results: Anterior pituitary function (GnRH stimulation test) correlated well with MRI results. However, no correlation was between the MRI measurements, the stimulation test and the clinical status of the patients, as 28 out of the 33 patients achieved normal puberty. Conclusions: MRI in conjunction with a GnRH stimulation test may be useful in predicting future impairment of pituitary function; however, further studies are needed to assess the effect of chelation therapy on the iron overload in the gland.

Berg D, Hoggenmuller U, Hofmann E, Fischer R, Kraus M, Scheurlen M, Becker G. 2000. The basal ganglia in haemochromatosis. Neuroradiology 42(1):9-13.
Abstract: Haemochromatosis is characterised by deposition of iron-containing pigment in various organs, but little is known about possible deposition in the brain and its clinical impact. We therefore investigated 14 patients with hereditary haemochromatosis with MRI, CT and transcranial ultrasound (TCS) and examined them neurologically. In six of the patients dense lesions were found within the lentiform nucleus on CT, all of whom displayed hyperechogenic lesions in the same area on TCS, as did one other patient. In these patients the relative signal intensities of the lentiform nucleus measured by MRI relaxometry were higher. No patient had clinical signs of basal ganglia disorders.

Barzilai A, Zilkha-Falb R, Daily D, Stern N, Offen D, Ziv I, Melamed E, Shirvan A. 2000. The molecular mechanism of dopamine-induced apoptosis: identification and characterization of genes that mediate dopamine toxicity. J Neural Transm Suppl (60):59-76.
Abstract: Parkinson's disease (PD) is a progressive neurological disorder caused by rather selective degeneration of the dopaminergic (DA) neurons in the substantia nigra. Though subject to intensive research, the etiology of this nigral neuronal loss is still enigmatic and treatment is basically symptomatic. The current major hypothesis suggests that nigral neuronal death in PD is due to excessive oxidative stress generated by auto- and enzymatic oxidation of the endogenous neurotransmitter dopamine (DA), the formation of neuromelanin and presence of high concentrations of iron. We have found that DA toxicity is mediated through its oxidative metabolites. Whereas thiol-containing antioxidants provided marked protection against DA toxicity, ascorbic acid accelerated DA-induced death. Using the differential display approach, we sought to isolate and characterize genes whose expression is altered in response to DA toxicity. We found an upregulation of the collapsin response mediator protein (CRM) and TCP-1delta in sympathetic neurons, which undergo dopamine-induced apoptosis. The isolation of these genes led us to examine the expression and activity of CRM and TCP-1delta related genes. Indeed, we found a significant induction of mRNAs of the secreted collapsin-1 and the mitochondrial stress protein HSP60. Antibodies directed against collapsin-1 provided marked and prolonged protection of several neuronal cell types

from dopamine-induced apoptosis. In a parallel study, using antisense technology, we found that inhibition of TCP-1delta expression significantly reduced DA-induced neuronal death. These findings suggest a functional role for collapsin-1 and TCP-1delta as positive mediators of DA-induced neuronal apoptosis.

Bartzokis G, Tishler TA. 2000. MRI evaluation of basal ganglia ferritin iron and neurotoxicity in Alzheimer's and Huntingon's disease. Cell Mol Biol 46(4): 821-833.
Abstract: Background: The basal ganglia contain the highest levels of iron in the brain and post-mortem studies indicate a disruption of iron metabolism in the basal ganglia of patients with neurodegenerative disorders such as Alzheimer's disease (AD) and Huntington's disease (HD). Iron can catalyze free radical reactions and may contribute to oxidative damage observed in ND and HD brain. Magnetic resonance imaging (MRI) can quantify transverse relaxation rates, which can be used to quantify tissue iron stores as well as evaluate increases in MR-visible water tan indicator of tissue damage). Methods: A magnetic resonance imaging (MRI) method termed the field dependent relaxation rate increase (FDRI) was employed which quantifies the iron content of ferritin molecules (ferritin iron) with specificity through the combined use of high and low field-strength MRI instruments. Three basal ganglia structures (caudate, putamen and globus pallidus) and one comparison region (frontal lobe white matter) were evaluated. Thirty-one patients with AD and a group of 68 older control subjects, and 11 patients with HD and a group of 27 adult controls participated (4 subjects overlap between AD and HD controls). Results: Compared to their respective normal control groups, increases in basal ganglia FDRI levels were seen in both AD and HD. FDRI levels were significantly increased in the caudate ($p = 0.007$) and putamen ($p = 0.008$) of patients with AD with a trend toward an increase in the globus pallidus ($p = 0.13$). In the patients with HD, all three basal ganglia regions showed highly significant FDRI increases ($p<0.001$) and the magnitude of the increases were 2 to 3 times larger than those observed in AD versus control group comparison. For both HD and AD subjects, the basal ganglia FDRI increase was not a generalized phenomenon, as frontal lobe white matter FDRI levels were decreased in HD ($p = 0.015$) and remained unchanged in AD. Significant low field relaxation rate decreases (suggestive of increased MR-visible water and indicative of tissue damage) were seen in the frontal lobe white matter of both HD and AD but only the HD basal ganglia showed such decreases. Conclusions: The data suggest that basal ganglia ferritin iron is increased in HD and PLD. Furthermore, the increased iron levels do not appear to be a byproduct of the illness itself since they seem to be present at the onset of the diseases, and thus may be considered a putative risk factor. Published post-mortem studies suggest that the increase in basal ganglia ferritin iron may occur through different mechanisms in HD and AD. Consistent with the known severe basal ganglia damage, only HD basal ganglia demonstrated significant decreases in low field relaxation rates. MRI can be used to dissect differences in tissue characteristics, such as ferritin iron and MR-visible water, and thus could help clarify neuropathologic processes in vivo. Interventions aimed at decreasing brain iron levels, as well as reducing the oxidative stress associated with increased iron levels, may offer novel ways to delay the rate of progression and possibly defer the onset of AD and HD.

Bartzokis G, Sultzer D, Cummings J, Holt LE, Hance DB, Henderson VW, Mintz J. 2000. In vivo evaluation of brain iron in Alzheimer disease using magnetic resonance imaging. Arch Gen Psychiatry 57(1):47-53.
Abstract: Background: The basal ganglia contain the highest levels of iron in the brain, and postmortem studies indicate a disruption of iron metabolism in the basal ganglia of patients with Alzheimer disease (AD). Iron can catalyze free radical reactions and may contribute to oxidative damage observed in AD brains. Treatments aimed at reducing oxidative damage have offered novel ways to delay the rate of progression and could possibly defer the onset of AD. Brain iron levels were quantified in

vivo using a new magnetic resonance imaging method. Methods: Thirty-one patients with AD and 68 control subjects participated in this study. A magnetic resonance imaging method was employed that quantifies the iron content of ferritin molecules (ferritin iron) with specificity through the combined use of high and low field-strength magnetic resonance imaging instruments. Three basal ganglia structures (caudate, putamen, and globus pallidus) and one comparison region (frontal lobe white matter) were evaluated. Results: Basal ganglia ferritin iron levels were significantly increased in the caudate (P = .007; effect size, 0.69) and putamen (P = .008; effect size, 0.67) of AD subjects, with a trend toward an increase in the globus pallidus (P = .13). The increased basal ganglia ferritin iron levels were not a generalized phenomenon; white matter ferritin iron levels were unchanged in patients with AD (P = .50). Conclusions: The data replicate and extend prior results and suggest that basal ganglia ferritin iron levels are increased in AD. Prospective studies are needed to evaluate whether premorbid iron levels are increased in individuals who develop AD.

Aschner M. 2000. Manganese: Brain transport and emerging research needs. Environ Health Perspect 108:429-432.
Abstract: Idiopathic Parkinson's disease (IPD) represents a common neurodegenerative disorder. An estimated 2% of the U.S. population, age 65 and older, develops IPD. The number of IPD patients will certainly increase over the next several decades as the baby-boomers gradually step into this high-risk age group, concomitant with the increase in the average life expectancy. While many studies have suggested that industrial chemicals and pesticides may underlie [PD, its etiology remains elusive. Among the toxic metals, the relationship between manganese intoxication and IPD has long been recognized. The neurological signs of manganism have received close attention because they resemble several clinical disorders collectively described as extrapyramidal motor system dysfunction, and in particular, IPD and dystonia. However, distinct dissimilarities between IPD and manganism are well established, and it remains to be determined whether Mn plays an etiologic role in IPD. It is particularly noteworthy that as a result of a recent court decision, methylcyclopentadienyl Mn tricarbonyl (MMT) is presently available in the United States and Canada for use in fuel, replacing lead as an antiknock additive. The impact of potential long-term exposure to low levels of MMT combustion products that may be present in emissions from automobiles has yet to be fully evaluated. Nevertheless, it should be pointed out that recent studies with Various environmental modeling approaches in the Montreal metropolitan (where MMT has been used for more than 10 years) suggest that airborne Mn revels were quite similar to those in areas where MMT was not used. These studies also show that Mn is emitted from the tail pipe of motor vehicles primarily as a mixture of manganese phosphate and manganese sulfate. This brief review characterizes the Mn speciation in the blood and the transport kinetics of Mn into the central nervous system, a critical step in the accumulation of Mn within the brain, outlines the potential susceptibility of selected populations (e.g., iron-deficient) to Mn exposure, and addresses future research needs for Mn.

Arlt S, Finckh B, Beisiegel U, Kontush A. 2000. Time-course of oxidation of lipids in human cerebrospinal fluid in vitro. Free Radic Res 32(2):103-114.
Abstract: Oxidative mechanisms play an important role in the pathogenesis of Alzheimer's disease, Parkinson's disease and other neurodegenerative diseases. To assess whether the oxidation of brain lipoproteins plays a role in the development of these pathologies; we investigated whether the lipoproteins of human cerebrospinal fluid (CSF) are susceptible to oxidative modification in vitro. We studied oxidation time-course for up to 100 h of human CSF in the absence (autooxidation) or presence of exogenous oxidants. Autooxidation of diluted CSF was found to result in a slow accumulation of lipid peroxidation products. The time-course of lipid hydroperoxide accumulation revealed three consecutive phases, lag-phase, propagation phase and plateau phase. Qualitatively similar time-course has been typically found in human plasma and plasma lipoproteins. Autooxidation of CSF was accelerated by adding exogenous oxidants,

delayed by adding antioxidants and completely inhibited by adding a chelator of transition metal ions. Autooxidation of CSF also resulted in the consumption of endogenous ascorbate, ol-tocopherol, urate and linoleic and arachidonic acids. Taking into account that (i) lipid peroxidation products measured in our study are known to be derived from fatty acids, and (ii) lipophilic antioxidants and fatty acids present in CSF are likely to be located in CSF lipoproteins, we conclude that lipoproteins of human CSF are modified in vitro during its autooxidation. This autooxidation appears to be catalyzed by transition metal ions, such as Cu(II) and Fe(III), which are present in native CSF. These data suggest that the oxidation of CSF lipoproteins might occur in vivo and play a role in the pathogenesis of neurodegenerative diseases.

Aoyama K, Matsubara K, Fujikawa Y, Nagahiro Y, Shimizu K, Umegae N, Hayase N, Shiono H, Kobayashi S. 2000. Nitration of manganese superoxide dismutase in cerebrospinal fluids is a marker for peroxynitrite-mediated oxidative stress in neurodegenerative diseases. Ann Neurol 47(4):524-527.
Abstract: Peroxynitrite can nitrate tyrosine residues of proteins. We examined nitrotyrosine-containing proteins in cerebrospinal fluid of 66 patients with neurogenic disease by immunoblot analysis. Nitrated tyrosine residue-containing protein was observed in the cerebrospinal fluid and was concluded to be manganese superoxide dismutase (Mn-SOD). The nitrated Mn-SOD level was strikingly elevated in amyotrophic lateral sclerosis patients and was slightly increased in Alzheimer's and Parkinson's disease patients, whereas an elevated Mn-SOD level was observed only in progressive supranuclear palsy group.

Ali MA, Yasui F, Matsugo S, Konishi T. 2000. The lactate-dependent enhancement of hydroxyl radical generation by the Fenton reaction. Free Radic Res 32 (5):429-438.
Abstract: The effect of lactic acid (lactate) on Fenton based hydroxyl radical ((OH)-O-.) production was studied by spin trapping, ESR, and fluorescence methods using DMPO and coumarin-3-carboxylic acid (3-CCA) as the (OH)-O-. traps respectively. The (OH)-O-. adduct formation was inhibited by lactate up to 0.4 mM (lactate/iron stoichiometry = 2) in both experiments, but markedly enhanced with increasing concentrations of lactate above this critical concentration. When the H2O2 dependence was examined, the DMPO-OH signal was increased linearly with H2O2 concentration up to 1 mM and then saturated in the absence of lactate. In the presence of lactate, however, the DMPO-OH signal was increased further with higher H2O2 concentration than 1 mM, and the saturation level was also increased dependent on lactate concentration. Spectroscopic studies revealed that lactate forms a stable colored complex with Fe3+ at lactate/Fe3+ stoichiometry of 2, and the complex formation was strictly related to the DMPO-OH formation. The complex formation did not promote the H2O2 mediated Fe3+ reduction. When the Fe3+-lactate (1:2) complex was reacted with H2O2, the initial rate of hydroxylated 3-CCA formation was linearly increased with H2O2 concentrations. All the data obtained in the present experiments suggested that the Fe3+-lactate (1:2) complex formed in the Fenton reaction system reacts directly with H2O2 to produce additional (OH)-O-. in the Fenton reaction by other mechanisms than lactate or lactate/Fe3+ mediated promotion of Fe3+/Fe2+ redox cycling.

Albin RL. 2000. Basal ganglia neurotoxins. Neurol Clin 18(3):665-+.
Abstract: The epidemiology, clinical features, pathology, and mechanisms of action of basal ganglia neurotoxins are reviewed. Manganese, cyanide, hydrogen sulfide, methanol, carbon monoxide, 3-nitropropionic acid, MPTP, and annonaceae alkaloids are discussed. The probable mechanism of action for almost all basal ganglia neurotoxins is inhibition of mitochondrial function with destruction of the pallidum and putamen. MPTP produces selective loss of dopaminergic neurons because of selective uptake of a toxic metabolite in dopaminergic neurons. The basis for selective vulnerability of the putamen and pallidum is unknown.

Al Moutaery K, Al Deeb S, Biary N, Morais C, Khan HA, Tariq M. 2000. Effect of

aluminum on neurological recovery in rats following spinal cord injury. J Neurosurg 93(2):276-282.
Abstract: Object. This investigation was undertaken to study the effect of aluminum on neurobehavioral, electrophysiological, structural, and biochemical changes in rats following spinal cord injury (SCI). Methods. Adult male Sprague-Dawley rats classified into different groups were given aluminum sulfate-dosed drinking water in the concentrations of 0%, 0.25%, 0.5% and 1%, respectively. After 30 days of aluminum treatment, the animals were subjected to spinal cord trauma. Laminectomy was performed at T7-8 in anesthetized rats, followed by placement of a compression plate (2.2 X 5 mm) loaded with a 35-g weight over the exposed spinal cord for 5 minutes. Control animals underwent the same surgical procedure, but the compression injury was not induced (sham). Postoperative neurological function was assessed using the inclined-plane test and by obtaining a modified Tarlov score and vocal/sensory score daily for 10 days. Electrophysiological changes were assessed using corticomotor evoked potentials, whereas pathological changes were assessed by light microscopy. The level of vitamin E in the: spinal cord was measured as an index of antioxidant defense. The behavioral, biochemical, and histological analyses were performed in a blinded fashion. Conclusions. Analysis of results obtained in the behavioral studies revealed that the compression of spinal cord produced transient paraparesis in which a maximum motor deficit occurred at Day I following SCI and resolved over a period of 10 days. Administration of aluminum significantly impaired the recovery following SCI. Analysis of the results of the biochemical, electrophysiological, and histopathological studies also confirmed the deleterious effects of aluminum on recovery from SCI in rats.

Aguilera M, Oliveros M, Martinez-Padron M, Barbas JA, Ferrus A. 2000. Ariadne-1: A vital drosphila gene is required in development and defines a new conserved family of RING-finger proteins. Genetics 155(3):1231-1244.
Abstract: We report the identification and functional characterization of ariadne-1 (ari-1), a novel and vital Drosophila gene required for the correct differentiation of most cell types in the adult organism. Also, we identify a sequence-related gene, ari-2 and the corresponding mouse and human homologues of both genes. All these sequences define a new protein family by the Acid-rich, RING finger, B-box, RING finger, coiled-coil (ARBRCC) motif string. In Drosophila, ari-1 is expressed throughout development in all tissues. The mutant phenotypes are most noticeable in cells that undergo a large and rapid membrane deposition, such as rewiring neurons during metamorphosis, large tubular muscles during adult myogenesis, and photoreceptors. Occasional survivors of null alleles exhibit reduced life span, motor impairments, and short and thin bristles. Single substitutions at key cysteines in each RING finger cause lethality with no survivors and a drastic reduction of rough endoplasmic reticulum that can be observed in the photoreceptors of mosaic eyes. In yeast two-hybrid assays, the protein ARI-1 interacts with a novel ubiquitin-conjugating enzyme, UbcD10, whose sequence is also reported here. The N-terminal RING-finger motif is necessary and sufficient to mediate this interaction. Mouse and fly homologues of both ARI proteins and the Ubc can substitute for each other in the yeast two-hybrid assay, indicating that ARI represents a conserved novel mechanism in development. In addition to ARI homologues, the RBR signature is also found in the Parkinson-disease-related protein Parkin adjacent to an ubiquitin-like domain, suggesting that the study of this mechanism could be relevant for human pathology.