## POSSIBLE ENVIRONMENTAL CAUSES OF ALTERED SEX RATIO

### Dioxin in Seveso, Italy

A striking illustration of the apparent ability of environmental exposures to influence male proportion comes from recent epidemiologic reports on the population most highly exposed to 2,3,7,8-tetrachlorodibenzodioxin (TCDD) from a chemical plant explosion in Seveso. In July 1976, an explosion released a large cloud dispersing many kilograms of this toxic agent into the atmosphere. A recent assessment of children born to the small number of highly exposed adults on whom data are available in this region found that for 7 years after this explosion, twice as many females as would be expected were born and overall fertility was markedly reduced.[31]

Between April 1977 and December 1984 (corresponding to 1 half-life of TCDD in the body), 74 children were born to parents in the zone of greatest exposure. Of these, 48 were female and 26 male, for a male proportion of 0.351. In 1976, serum samples were taken from the exposed populations; based on an assay (that was not available at the time of the explosion), it has been determined that for 7 years after the explosion, no boys were born to parents with the highest levels of dioxin in their blood. The proportion of female offspring proved to be highest among parents with the highest levels of serum TCDD. In fact, none of the 9 couples with the highest serum TCDD levels (at least 100 parts per trillion) bore a single male child.

Since 1985, the male proportion in this population has returned to expected levels and overall fertility has increased. This resumption of a normal pattern in the Seveso population further strengthens the argument that unusual environmental exposures can be the primary cause of reduced sex ratio in some circumstances.

### Other Possible Environmental Causes

Several researchers have assessed whether general environmental exposures to materials linked to alterations in sex at birth in highly exposed workers might also produce alterations in children conceived in neighborhoods with similar exposures.[32] The evidence on this matter remains incomplete, but provocative. Five different retrospective studies of heavily polluted Scottish residential areas revealed significantly impaired sex ratio.[33] The pollutants included some highly visible emissions from acrid smelters, steel foundries, and incinerators in Scotland between 1975 and 1983.

## INCREASED PRENATAL VULNERABILITY OF THE MALE

There are compelling biological reasons for deducing that whatever factors may be altering phenotypic sex at birth could also be involved in producing other adverse effects in males. Disorders of the reproductive tract, such as hypospadias and cryptorchidism, are reported to have increased in many industrial countries.[34] A recent analysis in the United States showed that the rate of hypospadias had nearly doubled in all 4 regions of the United States from 1970 to 1993.[35] During this same period, testicular cancer rates have also increased.[36] A recent study reports a link between exposure to polyvinyl chloride and testicular cancer.[37] A number of reports have recently reviewed this evidence and speculated that these phenomena are biologically connected to critical exposures to xenohormones that occur immediately before and early in the process of reproduction.[38,39]

Both human and animal evidence demonstrate that prenatal exposures may be far more important to the overall health and development of the organism than those that occur later in life.[40] Thus, exposures during the period of organogenesis and differentiation of the genitals during weeks 6 and 9 chemically imprint rapidly growing reproductive tract cells for later developmental disorders, ranging from testicular cancer to reproductive difficulties.[41]

Recent innovative analysis of geographic patterns of cryptorchidism suggests that parental exposure to hormone-disruptive chemicals increases the risk of this disorder.[42] Cryptorchidism is a birth defect of the male reproductive tract that is typically corrected surgically through cryptorchidopexy. This study compared rates of male reproductive tract defects in different regions of the Spanish province of Granada. Although fruit and vegetable crops in this region take up only 4.65% of Spain's farmland, they are treated with 51% of the pesticides used in the country. In much of the area along the Mediterranean coast, greenhouse crop farming under plastic-encased systems is widespread. In the enclosed greenhouses, workers (including pregnant women) are exposed to high levels of pesticides, many of them organochlorine compounds that have been shown to damage the endocrine system. When districts within Granada were categorized based on pesticide use, it was found that rates of cryptorchidopexy in the population were highest in those districts where pesticide use was high. In these areas, the level of cases was 2.32 times the rate in areas not exposed to pesticides ($P<.05$).

Further evidence of a link between defects in male reproductive outcomes and reduced male proportion is provided by recent observations from rural Minnesota.[43] Increased rates of birth defects have occurred in male children of workers who apply pesticides, suggesting that the developing male fetus may be especially vulnerable to hormone-disrupting substances. Compared to the general population, children born to workers who apply pesticides, herbicides, and fungicides were 1.57 times as likely to have birth defects of the circulatory and/or respiratory, urogenital, and musculoskeletal and/or integumental systems. In agricultural areas, increased rates of birth defects were also seen in the general population. For example, in western Minnesota where spring wheat production predominates and fungicides are aerially applied, 2.6% of live births showed anomalies, as compared with 1.83% of births in noncrop regions. The increase was especially pronounced for infants conceived in the spring, when chlorophenoxy herbicides were routinely applied.

Interestingly, more male than female infants were affected by birth defects in Minnesota regions with elevated rates of use of some herbicides and fungicides. Among all births with anomalies, the proportion of males was 0.579. In areas of high use of chlorophenoxy herbicides and/or fungicides, the male proportion among children born with defects to workers who apply pesticide was 0.736, compared with 0.607 for births with anomalies in the general population. In areas of low pesticide usage, the male proportion born with defects was 0.682 for children of fathers who were pesticide appliers compared with 0.633 for the general population. Thus, it appears that the male fetus is more vulnerable to paternal exposures that take place prior to conception and that may be linked with birth defects.[2]

## COMMENT

With respect to the quality of data and record keeping on sex at birth, all of the countries reported here have no known cultural biases regarding reporting or selection of sex through elective abortion. This suggests that recorded trends in sex at birth are likely to reflect real patterns rather than artifacts of measurement or cultural bias.

The ratio of human males to human females at birth is not static. Between 1900 and 1950, the proportion of males rose significantly in several industrial countries, beyond the range of historical variation. This was chiefly due to general improvements in obstetrical care, which produced a decline in stillbirths,[7,44,45] that

disproportionately affect males. In general, male fetuses appear to be more susceptible to reproductive hazards, as they also experience higher rates of many birth defects than do females. Since 1970, a significant downward trend has occurred in the proportion of males in the Netherlands, Denmark, Canada, and the United States, and recent reports indicate that parallel declines also have occurred in Sweden, Germany, Norway, and Finland.[19] It is distinctly possible that the causes of these trends, however, are not benign. Rather, the reduction of the proportion of males born may be a sentinel health event that some, as yet, unrecognized environmental health hazards are affecting the sex ratio of births as well as other unexplained defects in male reproduction.

Provocative theories about shifting parental hormonal milieus have been devised to account for some reported alterations in sex at birth. Some observers have speculated that increases in male proportion after major wars are due to an increase in coital frequency.[20] Increased sexual activity appears to result in earlier fertilizations within the cycle and in more males being conceived and born.[17] The recent decline in the proportion of male births has been explained by some as a return to normal after a wartime high. However, our analysis indicates that this proportion continues to decline several decades after the wartime high, suggesting that other factors must be contributing to the trends.

Recent trends and geographic patterns in sex ratio cannot be attributed to documented causes of altered sex ratio, such as physiological conditions and medical treatments. Known causes of altered sex ratio, such as older age of father, use of in vitro fertilization, and stress of mothers, appear unlikely to account for trends that have persisted throughout the past 2 decades. In fact, the decline in Canada is greatest in those regions where known medical causes of reduced male proportion such as in vitro fertilization are likely to be lowest. In Canada overall, in vitro fertilization only accounts for some fraction of the recent reductions in male births each year.[10] Infertility affects about 7% to 8% of all couples. Based on survey data, it appears that about 8% of infertile couples undergo ovulation induction. If about 25% of these succeed in producing live births, far fewer than 1% of live births annually in Canada could be due to ovulation induction. If induction decreases the proportion of male births to 46%, then as many as 350 fewer male births would result annually in recent years. But, this explanation cannot account for the overall declines in male proportion observed in the 1970s and early 1980s, when in vitro fertilization was not widely available.

The study of sex determination remains a field full of speculation and with limited empirical evidence. As a consequence, factors that affect the sex ratio remain poorly understood. Many of the causes of reduced male births that have been identified, such as stress of fathers, in vitro fertilization, less frequent intercourse, and multiple sclerosis, are unlikely to account for the time trends that have recently been observed in several industrial countries. Several specific workplace and environmental exposures have altered the sex ratio in those who were highly exposed to some pesticides and other general environmental contaminants. Whether these agents could account for some of the recently observed patterns is a matter of considerable concern.

## CONCLUSIONS

We propose that reduced male proportion at birth be viewed as a sentinel health event that may be linked to environmental factors. To determine the value of this suggestion, it will be important to answer a number of questions: Do the trends in sex ratio reported for the United States, Canada, Denmark, and the Netherlands parallel similar changes in other countries? Are regional differences in the proportion of males consistent with environmental factors or other known causes of alterations? Does other evidence confirm that occupational cohorts with exposures to smelting operations, pesticides, inorganic borates, lead, solvents, alcohol, and other such workplace hazards have produced children with reduced male proportion?

To resolve these matters, it will be important for public health researchers to conduct a number of assessments, examining patterns, and time series of state, regional, and national birth registries. Information on sex ratio has the virtue of being easy to obtain in countries with limited resources for health and environmental research. In contrast to studies of morbidity, sex ratio is not susceptible to diagnostic bias or changes in ascertainment. Geographic monitoring of changes in sex ratio could prove a useful tool for assessing whether specific, avoidable medical or environmental exposures are occurring in specific regions.

Relatively small reductions in male proportion over the past 2 decades in the United States and Canada theoretically account for decreases in about 38 000 and 8600 male births, respectively.[8] The potential repercussions of conditions that may alter the ratio of the sexes at birth should be considered a matter of utmost concern. The extent to which other adverse health consequences are linked to this phenomenon is a matter of grave importance for public health.

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