

Bisphenol A Eliminates Brain and Behavior Sex Dimorphisms in Mice: How Low Can You Go?

In this issue, Rubin and colleagues (1) report on neurobehavioral effects caused by developmental exposure in mice to minute doses of the ubiquitous environmental chemical bisphenol A (BPA). BPA is one of the highest-volume chemicals and is produced in excess of 6 billion pounds per year for use as the monomer that is polymerized to manufacture polycarbonate plastic food and beverage containers, the resin lining of metal cans, dental sealants, and as an additive in a wide array of other products. BPA was reported by Dodds and Lawson (2) in 1936 to be a full estrogen agonist, before chemical engineers determined in the 1950s that this hormonally active drug could be polymerized to produce polycarbonate plastic. Limited research on BPA conducted by toxicologists (3) led to the proposal that a dose of 50 mg/kg·d was the no-effect dose (4). However, the initial toxicological studies did not take into account that the approach to studying the effects of BPA in animals should be based on methods appropriate for examining an estrogenic drug rather than on standard toxicological methods, where only a few very high doses based on acute toxic effects are examined and a “safe” dose for daily human exposure is estimated but never directly tested (5). The findings reported here and in previous studies by Rubin and colleagues (5) are stunning in that they reveal permanent effects on brain morphology, neurochemistry, and behavior in mice at doses 200,000 to 2 million times lower than the daily dose that chemical corporations proposed caused no effect only a decade ago, which was used to estimate the daily dose currently accepted as safe by the Food and Drug Administration (FDA).

The approach to administering BPA by Rubin *et al.* (1) is markedly different from the typical approach in toxicological research of a gavage administration once per day. Rubin *et al.* (1) chose to use a method of administration (sc implanted Alzet osmotic pump) that mimics the continuous exposure to BPA that extensive research suggests is occurring in human populations, and the doses administered are clearly relevant to human exposure levels. Specifically, over the last 6 yr, approximately 20 published studies have reported that unconjugated (parent) BPA is detected in virtually everyone in the United States, Europe, and Asia who is examined, always within the range of approximately 0.1–10 ng/ml (median and mean of ~10 nM) in both blood and tissue, and within the same range in urine after conjugation. BPA shows very limited binding to estrogen-binding plasma proteins in humans and rodents, and BPA alters human, rat, and mouse cell function at doses as low as 1 pM, far below its free level in human blood (6). BPA is rapidly metabolized, and the ab-

sence of a significant proportion of people without detectable BPA within a bioactive range suggests that continuous human exposure to this ubiquitous chemical must be occurring throughout the world. This is not unexpected, because hydrolysis of the ester bond linking BPA molecules in polycarbonate plastic and resins increases as a function of temperature and either high or low pH (Fig. 1).

It is well established that estrogens mediate various aspects of sexual differentiation, although species vary with regard to the specific aspects of development that are directly or indirectly affected by variation in estrogen exposure during critical periods in development. Previous studies from this group and others over the past 15 yr have revealed that there are numerous environmental chemicals, in addition to BPA, that mimic the activity of estradiol and thus have the potential to interact in disrupting development of estrogen-responsive tissues. The constant characterization of all of these environmental chemicals as weak in industry-sponsored publications has led to the general belief, referred to as manufactured doubt (7), that human exposure to low doses of chemicals such as BPA is not a public health concern (5). The findings presented by Rubin *et al.* (1) strongly argue for a greater level of concern, particularly because, unlike drugs, interactive effects of environmental chemicals that operate through common mechanisms are not considered in assessing the potential risks to human health posed by these chemicals.

An area of the developing rodent brain that is well characterized as being responsive to estrogens is the region of the preoptic area (the anteroventral periventricular preoptic area) examined by Rubin *et al.* (1). This area of the brain is involved in the regulation of gonadotropin secretion as well as many aspects of social and sexual behaviors. In previous studies, Rubin, Soto, and their colleagues have reported that exposure via the pregnant and lactating dam to the extremely low doses used in the present study (0.025 and 0.25 $\mu\text{g}/\text{kg}\cdot\text{d}$) disrupt the rate of postnatal growth, timing of puberty, and subsequent estrous cyclicity as well as uterine and mammary gland morphology and function in female mice. These findings have been independently confirmed by other investigators using other animal models and other methods of administration of low doses of BPA (8).

A particularly interesting aspect of the findings by Rubin *et al.* (1) is that the effect of BPA on the number of neurons in the anteroventral periventricular preoptic area that express tyrosine hydroxylase was to eliminate the sexual dimorphism seen in control males and females, with BPA-exposed female offspring showing a decrease to control male levels. The finding that developmental exposure to low doses of BPA eliminates or even reverses sex differences in brain morphology, function, and behavior is consistent with a number of other reports (8). Finally, although Rubin and

Abbreviation: BPA, Bisphenol A.

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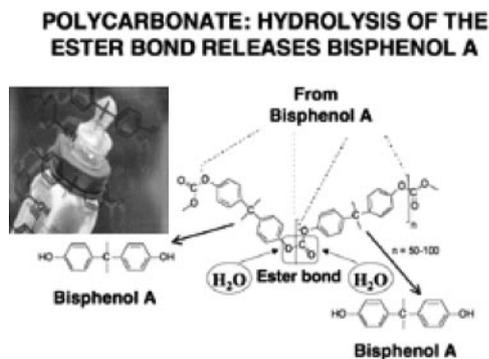


FIG. 1. Schematic diagram depicting hydrolysis of the ester bond linking BPA molecules in polycarbonate plastic (such as the baby bottle shown in the *inset*) as well as resins used to line metal cans. The ester bond is subject to an increase in hydrolysis as temperature increases and in response to acidic or basic conditions.

colleagues have focused in their previous studies on neuroendocrine effects directly related to regions of the preoptic area, in the present study, they chose to examine a number of measures of activity that are sexually dimorphic in adults. Here they add to a growing literature that developmental exposure to BPA permanently alters activity levels, and as was the case for the tyrosine hydroxylase results, once again the effect was to eliminate the sexual dimorphism observed in unexposed animals. Taken together, these findings suggest very interesting differences between males and females in the neurobehavioral impact of exposure to extremely low doses of BPA and thus likely also other estrogenic chemicals. The extremely low doses of BPA that caused these neurobehavioral changes in mice are far below the acceptable daily intake considered “safe” for humans by the FDA. Together with the human relevance of the method of administration used in this study, this should stimulate the FDA to recon-

sider its current position that it is unconcerned with the levels of BPA to which everyone in the United States (and elsewhere in the world) are exposed.

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