Environmental toxins

Exposure to bisphenol A advances puberty

Plastics and pesticides are examples of products that contain oestrogenic endocrine-disrupting chemicals, or EEDCs, which can interfere with mammalian development by mimicking the action of the sex hormone oestradiol[1]. For instance, the exposure of developing rodents to high doses of EEDCs advances puberty and alters their reproductive function[2]. Low environmental doses of EEDCs may also affect development in humans[3]. Effects have become apparent in humans over the past half century that are consistent with those seen in animals after exposure to high doses of EEDCs, such as an increase in genital abnormality in boys[4] and earlier sexual maturation in girls[5]. Here we show that exposing female mouse fetuses to an EEDC at a dose that is within the range typical of the environmental exposure of humans alters the postnatal growth rate and brings on early puberty in these mice.

Oestrogen is a hormone that interacts with other steroids to regulate the normal development of the reproductive system and other tissues. Bisphenol A, a compound that was initially synthesized as a chemical oestrogen[6], is now used as the monomer for the production of polycarbonate plastic products such as baby bottles. Bisphenol A leaches out of such products at a rate that increases with repeated use[7].

Pregnant CF-1 mice ($n=21$ per treatment group) were fed either oil (vehicle) or bisphenol A dissolved in oil at a dose equivalent to that typically found in the environment (2.4 μg per kg), on days 11 to 17 of gestation[8]. Pups were delivered by caesarean section on day 19 to determine their position in the uterus and were reared by untreated foster mothers. The intrauterine position determines fetal hormone levels because endogenous sex steroids are transported from one fetus to another[9]. Mouse fetuses positioned between two males (Fig. 1, 2M) are exposed to the lowest levels of oestradiol, fetuses located next to female fetuses (0M) are exposed to the highest, and females next to one male (1M) are exposed to an intermediate amount[10].

At weaning on postnatal day 22, females treated with bisphenol A were significantly heavier than control females (Fig. 1a), although they had a similar body weight at birth: relative to controls from the same intrauterine position, the weight of 0M females was increased by 22% and that of 1M females was increased by 9%; 2M females were unaffected (Fig. 1d). The findings were virtually identical for male siblings.

On postnatal day 26, females were housed individually but near to males to provide a submaximal level of pheromonal stimulation[11]. After vaginal opening, daily vaginal smears were examined for the presence of completely cornified epithelial cells (a sign of first vaginal oestrus). We found that prenatal treatment with bisphenol A significantly reduced the number of days between vaginal opening and first vaginal oestrus, which is highly correlated with first postpubertal ovulation[12] (Fig. 1c), in 0M females but not in 2M females (Fig. 1f), based on analysis of covariance adjusted for body weight at weaning.

Prenatal exposure to a dose of bisphenol A comparable to levels found in the environment therefore altered postnatal growth rate and reproductive function in female mice, although individual differences in endogenous oestradiol resulting from natural variation influenced the responsiveness of the females to bisphenol A.

There is significant variability in human and animal populations in responsiveness

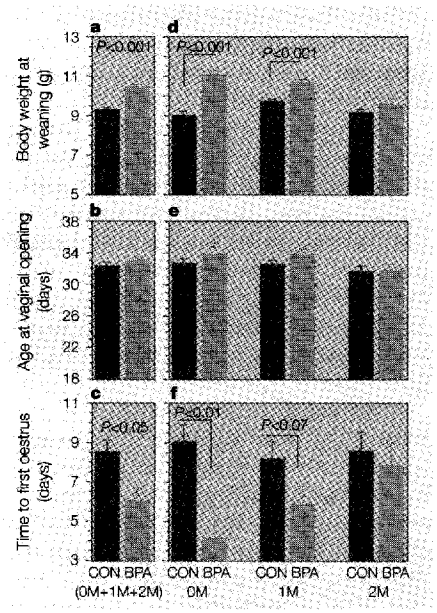

Figure 1 Mean (±s.e.m.) body weight at weaning, age at vaginal opening, and interval between vaginal opening and first vaginal oestrus. Data are for all females combined (**a–c**) and as a function of intrauterine position (**d–f**). Body weight at weaning includes data for all females surviving to weaning from control (CON, orange bars) and bisphenol A-exposed (BPA, green bars) litters. All data were adjusted for litter membership to control for maternal effects. Vaginal opening and interval data were also corrected by analysis of covariance for body weight at weaning. 2M, located between two males fetuses; 1M, located next to one male fetus; 0M, located next to female fetuses. Wean weight was calculated on 41 0M, 47 1M and 23 2M control females, and 20 0M, 43 1M and 12 2M bisphenol A-treated females. Vaginal opening and interval data were calculated on 19 0M, 20 1M and 19 2M control females, and on 19 0M, 21 1M and 11 2M bisphenol A-treated females. We attempted to include females from each intrauterine position from each litter, but some litters did not contain a 2M female.

to hormones and EEDCs. Our findings indicate that one source of this variability may be the amount of endogenous sex hormones. These vary among individual human fetuses and are influenced by a variety of factors, including whether it is a first pregnancy, the size of the placenta, and whether there is just one fetus or twins[13,14].

Very small increases in the level of endogenous oestradiol may substantially increase the sensitivity of fetuses to EEDCs consumed by pregnant women, so some fetuses may be at particularly high risk for a wide array of abnormalities and diseases. Our findings emphasize the need for studies to examine the relationship between maternal exposure to endocrine disrupters and subsequent health effects in the offspring.

Kembra L. Howdeshell*, Andrew K. Hotchkiss†, Kristina A. Thayer*, John G. Vandenbergh†, Frederick S. vom Saal*

**Division of Biological Sciences, University of Missouri, Columbia, Missouri 65211, USA*
e-mail: vomsaalf@missouri.edu
†Department of Zoology, North Carolina State University, Raleigh, North Carolina 27695-7617, USA

1. Colborn, T., vom Saal, F. S. & Soto, A. M. *Environ. Health Perspect.* **101,** 378–384 (1993).
2. Gray, L. E. Jr. in *Chemically Induced Alterations in Sexual and Functional Development* Vol. 21, *The Wildlife/Human Connection* (eds Colborn, T. & Clement, C.) 203–230 (Princeton Scientific, New Jersey, 1992).
3. vom Saal, F. S. & Sheehan, D. M. *Forum* **13,** 11–18 (1998).
4. Paulozzi, L. J., Erickson, J. D. & Jackson, R. J. *Pediatrics* **100,** 831–834 (1997).
5. Herman-Giddens, M. E. *et al. Pediatrics* **99,** 505–512 (1997).
6. Dodds, E. C. & Lawson, W. *Nature* **137,** 996 (1936).
7. Takao, Y. *et al. J. Health Sci.* **45,** 39 (1999).
8. Nagel, S. C. *et al. Environ. Health Perspect.* **105,** 70–76 (1997).
9. Even, M. D., Dhar, M. G. & vom Saal, F. S. *J. Reprod. Fertil.* **96,** 709–716 (1992).
10. vom Saal, F. S. *J. Anim. Sci.* **67,** 1824–1840 (1989).
11. Vandenbergh, J. G. *Endocrinology* **84,** 658–660 (1969).
12. vom Saal, F. S., Pryor, S. & Bronson, F. H. *J. Reprod. Fertil.* **62,** 33–37 (1981).
13. Panagiotopoulou, K. *et al. Cancer Causes Control* **1,** 119–124 (1990).
14. Hsieh, C.-C. *et al. Am. J. Epidemiol.* **136,** 1321–1326 (1992).