

Commercial Animal Feed: Variability in Estrogenic Activity and Effects on Body Weight in Mice

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Mammalian embryonic development is epigenetic in that hormonal signals control not only the timing of gene expression but set the activity of genes and thus the functioning of organs and homeostatic systems for the remainder of life (Newbold et al., 2004). Variation in endogenous hormones, such as estradiol and testosterone, which regulate development of organs (vom Saal, 1989), or disruption of the activity of these hormones by chemicals during development can lead to permanent changes in homeostatic systems, such as the regulation of fat and body weight. These changes can have profound effects on a variety of diseases (Barker et al., 1993).

There is evidence that during postnatal life, estradiol has an inhibitory effect on body fat, because ovariectomy results in an increase in body weight in females; mice without the alpha form of estrogen receptors are heavier than mice with normal estrogen receptors (Heine et al., 2000). However, different effects can occur when exposure to elevated levels of estradiol or other estrogenic chemicals occurs during fetal development. Newbold (this symposium) has reported that female mice exposed to low levels of the drug diethylstilbestrol during fetal life eventually become fatter than nonexposed mice. We and others have reported that fetal exposure to very low doses of the estrogenic chemical bisphenol A, which is the monomer used to make polycarbonate plastic, results in an increase in postnatal growth (Howdeshell et al., 1999; Rubin et al., 2001). Related to this finding is the report that bisphenol A at 2 µg/ml accelerated the conversion of mouse 3T3-L1 fibroblast cells into adipocytes and also increased lipoprotein lipase activity and triacylglycerol accumulation. Over time, bisphenol A resulted in the presence of larger lipid droplets in the differentiated cells. Insulin and bisphenol A interacted synergistically to further accelerate these processes (Masuno et al., 2002). In a related study, bisphenol A stimulated an increase in the glucose transporter (GLUT-4) and glucose uptake into 3T3-F442A adipocytes in cell culture. Of interest, this effect of bisphenol A was not inhibited by the estrogen receptor antagonist ICI 182,780, revealing that this effect is not mediated by nuclear estrogen receptors (Sakurai et al., 2004). Another related finding is that in pancreatic β cells, bisphenol A stimulates phosphorylation and activation of the transcription factor CREB within a few minutes via a mechanism that is dependent on the rapid influx of calcium into the cell rather than binding to nuclear estrogen receptors. CREB plays an important role in adipocyte differentiation, and it is possible that the effects of bisphenol A on adipocyte differentiation thus also involve rapid effects on CREB.

Not all estrogenic chemicals show the same spectrum of effects in target tissues. This property of estrogenic chemicals has led to the description of these chemicals as selective estrogen receptor modulators or SERMs. Thus, while bisphenol A has some effects that are similar to those of estradiol, other effects, such as those described above in adipocytes, are not similar to the effects of estradiol. In fact, we have evidence that another class of estrogenic chemicals, namely those that are present in commercial animal feeds used in laboratory experiments, can lead to a reduction in body fat rather than a stimulation of body fat (vom Saal, unpublished observation). Specifically, as the amount of estrogenic contaminants in some batches of a casein-based animal feed increased, the amount of body fat decreased in mice maintained on this feed throughout life. One issue here that is of considerable concern is that commercial feeds used in laboratory experiments with rodents are not routinely screened for levels of contaminants with estrogenic activity. In addition to naturally occurring phytoestrogens, such contaminants can include mycotoxins, such as zeralonone, which is a potent estrogenic chemical.

The finding that different types of chemicals classified as “estrogens,” due to their ability to interact with nuclear estrogen receptors, exert different types of activities in certain target tissues should not be viewed as surprising or contradictory. The above findings with bisphenol A show that these chemicals can exert effects through many different pathways. In addition, after binding to either the alpha or beta form of estrogen receptors, these chemicals are known to exert unique effects depending on coregulators that are part of the transcriptional apparatus for steroid hormone receptors (Routhledge et al., 2000).

A final issue of interest is that estrogenic chemicals can impact nursing performance in mammals. Estrogen inhibits the action of prolactin in mammary gland epithelium, thus reducing lactation. We have found that as the amount of estrogenic activity in animal feed increases, the amount of time spent nursing decreases in lactating mice (Giosa and vom Saal, unpublished observation). We also have reported that exposure to bisphenol A only during pregnancy results in a subsequent reduction in nursing behav-

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ior in mice (Palanza et al., 2002), which is associated with a decrease, rather than increase, in the rate of growth of offspring (Honma et al., 2002).

Clearly, the effects of estrogen on the regulation of fat and body weight are complex. The potential for unique developmental effects due to exposure during critical periods in tissue differentiation needs to be considered. In addition, the potential for unique effects of different chemicals classified as "environmental estrogens" also has to be taken into account. Finally, the actions of estrogenic chemicals on the brain and other physiological processes, such as lactation, have to be considered in order to understand effects of developmental exposure on growth and subsequent body composition and size.

REFERENCES

- Barker DJ, Gluckman PD, Godfrey KM, et al. 1993. Fetal nutrition and cardiovascular disease in adult life. *Lancet* 341:938–941.
- Heine PA, Taylor JA, Iwamoto GA, et al. 2000. Increased adipose tissue in male and female estrogen receptor-alpha knockout mice. *Proc Natl Acad Sci USA* 97:12729–12734.
- Honma S, Suzuki A, Buchanan DL, et al. 2002. Low dose effect of in utero exposure to bisphenol A and diethylstilbestrol on female mouse reproduction. *Reprod Toxicol* 16:117–122.
- Howdeshell KL, Hotchkiss AK, Thayer KA, et al. 1999. Exposure to bisphenol A advances puberty. *Nature* 401:763–764.
- Masuno H, Kidani T, Sekiya K, et al. 2002. Bisphenol A in combination with insulin can accelerate the conversion of 3T3-L1 fibroblasts to adipocytes. *J Lipid Res* 43:676–684.
- Newbold RR, Jefferson WN, Padilla-Banks E, Haseman J. 2004. Developmental exposure to diethylstilbestrol (DES) alters uterine response to estrogens in prepubescent mice: low versus high dose effects. *Reprod Toxicol* 18:399–406.
- Palanza P, Howdeshell KL, Parmigiani S, vom Saal FS. 2002. Exposure to a low dose of bisphenol A during fetal life or in adulthood alters maternal behavior in mice. *Environ Health Perspect* 110:415–422.
- Routhledge EJ, White R, Parker MG, Sumpter JP. 2000. Differential effects of xenoestrogens on coactivator recruitment by estrogen receptor (ER) α and ER β . *J Biol Chem* 275:35986–35993.
- Rubin BS, Murray MK, Bamassa DA, et al. 2001. Perinatal exposure to low doses of bisphenol A affects body weight, patterns of estrous cyclicity, and plasma LH levels. *Environ Health Perspect* 109:657–680.
- Sakurai K, Kawazuma M, Adachi T, et al. 2004. Bisphenol A affects glucose transport in mouse 3T3-F442A adipocytes. *Br J Pharmacol* 141:209–214.
- vom Saal FS. 1989. Sexual differentiation in litter-bearing mammals: influence of sex of adjacent fetuses in utero. *J Anim Sci* 67:1824–1840.