DEVELOPMENT AND HEPATIC ENZYMES ACTIVITIES IN FEMALE MICE PRENATALLY EXPOSED TO DECABROMINATED DIPHENYL ETHER (PBDE 209)

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Introduction

Decabrominated diphenyl ether (PBDE 209) is mostly used as flame retardant additives to electronic enclosures such as television set cabinet backs and in upholstery textiles.[1] It is additives therefore may be released into the environment when the products are manufactured, used or disposed.[2] Some studies found a high association between maternal and fetal cord blood PBDE concentrations[3,4] as well as eggs sample were detected PBDEs (including PBDE 209).[5,6] These study show that PBDEs might be able to enter the fetus through the placenta. PBDEs structural similarities persistent aromatic compounds, suggest that PBDEs might activate the aryl hydrocarbon receptor (AhR) signal transduction pathway.[7] This response leads to the induction of the cytochrome P-450 isozyme CYP 1A1. Many studies investigated the congeners 99 and the penta-BDE mixture DE-71 have been demonstrated to induce both phase I [ethoxyresorufin-O-deethylase (EROD) and pentoxyresorufin-O-deethylase (PROD)], and phase II metabolic enzyme activity [uridinediphosphate-glucuronosyltransferase (UDPGT)] in prenatal exposure.[8,9] A few toxicological studies have been carried out and the PBDE 209 can elicit hepatic enzymes activities *in vivo* and has an endocrine disruptor potential in male offspring mice.[10] However, little is known about the developmental toxicity of PBDE 209 in females. The objective of the present study was to assess whether prenatal exposure to PBDE 209 affects the development and hepatic enzyme levels in dams and female offspring mice.

Materials and Methods

Animals and treatment. Pregnant ICR outbred mice were randomly divided into four groups of five mice each and housed individually. Dams were gavaged daily with corn oil (control), 10, 500 and 1500 mg/kg bw PBDE 209 in corn oil from gestation days 0-17. During the pup's lactation period, developmental landmarks including pinnae detachment, body hair fuzz appearance, incisor eruption, ear opening, and eyes opening day were recorded. After weaning, five dams from each treatment group were euthanized. The livers supernatant (S9) samples were preserved at -80℃ until hepatic enzyme activities analysis. On postnatal day (PND) 21, 35, and 70 that two or three female offspring per litter were randomly selected. The animal was killed and their organs including liver, kidney, adrenal, spleen, and ovary were dissected and weighed. Liver supernatant (S9) samples were preserved at -80℃ except offspring of PND 70.

Prepared of liver supernatant (S9) samples and Hepatic enzyme Assays. Liver S9 fractions were prepared as described previously.[10] Briefly, livers were perfused *in situ* with ice-cold 0.05 M Tris–0.15 M KCl buffer (pH 7.4) and weighed. The livers were then homogenized in the same Tris-KCl buffer. The crude homogenate was centrifuged at 10,000 × *g* for 15 min at 4°C. The pellet from the final centrifugation was resuspended in buffer (0.05 M Tris-HCl, 20% glycerol [v/v], 1 mM EDTA; pH 7.4 at 4°C) and stored at -80°C until assayed. Analyses of EROD and UDPGT were based on other previous studies.[10] Protein content was determined by the Bradford method using bovine serum albumin as a standard.[11]

Data analysis. Data were expressed as means ± standard error mean (SEM). All statistical analyses were performed on JMP 5.0. For the analysis of female offspring, the litter was considered the experimental unit. A p value of < 0.05 was considered statistically significant.

Results and Discussion

Effects on the development landmarks of female offspring mice. In dams treated with PBDE 209, we observed no significant developmental delays in any of development landmarks in control and PBDE 209-treated pups (data not shown).

organ weights. We found no significant differences in the relative weights of liver, kidney, adrenal glands, spleen, and ovaries, compared to the controls on PND 21, 35, and 70 (data not shown)..

Liver Enzymes. Liver wet weights of female offspring were no significantly different among different treatment groups. No significant changes were observed in hepatic enzyme activity of S9 EROD in any of the treatment groups on PND 21 and PND 35 (Fig. 1 and 2A). In addition, no significant effect were observed in hepatic enzyme activity of and S9 UDPGT in any of the treatment groups on PND 35 (Fig. 2B). Both EROD or UDPGT activities showed no dose-related increases for female offspring. However, liver S9 EROD activity was significant 21-fold increase of dam in highest dose group compared to control animals (Fig. 3A). Moreover, hepatic enzyme activity of S9 UDPGT was found in the 10- mg/kg/day groups that has a significant decrease ($P < 0.01$) compare with other group (Fig. 3B). Interesting, the results different from our study in the postnatal exposure to PBDE 209 on male mice that liver S9 EROD and 4-nitrophenol UDPGT activities were not significantly increased in PBDE 209-treated male mice. Our finding suggested that pregnant exposure to PBDE 209 might induce hepatic enzymes activities. We believed that the results will be important to the issue of environmental pollution and provide the critical information for pregnant health risk in the future.

Acknowledgements

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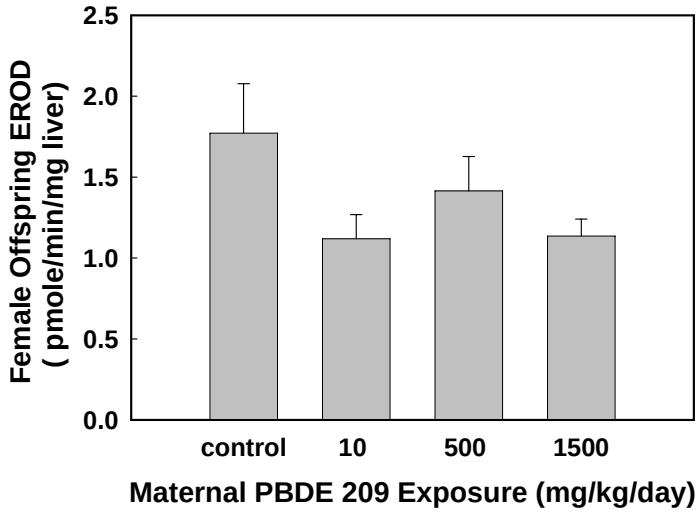

Fig. 1. S9 7-ethyoxyresorufin O-deethylase (EROD) activity in female offspring prenatally exposed to decabrominated diphenyl ether (PBDE 209) on postnatal day (PND) 21. Data are presented as means ± SEM.

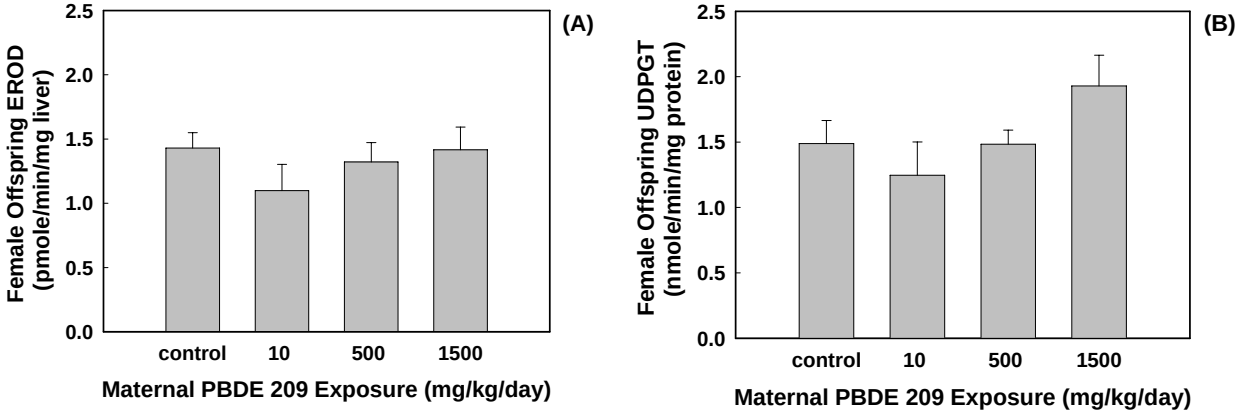

Fig. 2. Lack of effect of prenatal exposure to decabrominated diphenyl ether (PBDE 209) and controls on liver enzyme activity, S9 4-nitorphenol 7-Ethyoxyresorufin O-deethylase (EROD) activity (A) and 4-nitrophenol uridinediphosphate-glucuronosyltransferase (UDGPT) activity (B) in female offspring mice on postnatal day (PND) 35. Data are presented as means ± SEM.

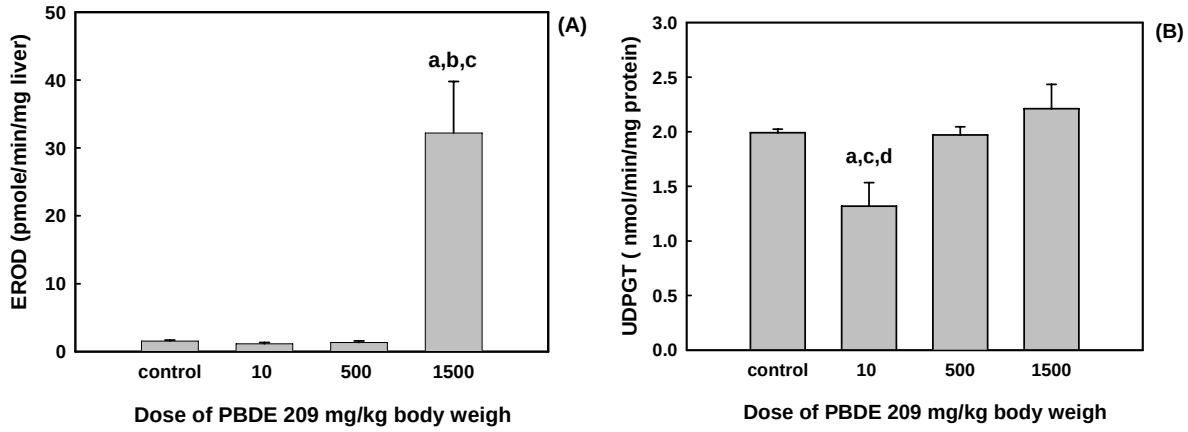

Fig. 3. The effect of pregnant mice exposure to PBDE 209 on liver enzyme activities, including S9 7-ethyoxyresorufin O-deethylase (EROD) activity (A) and 4-nitrophenol uridinediphosphate-glucuronosyltransferase (UDGPT) activity (B) after the period of lactation. Data are presented as means ± SEM. (a) $p < 0.01$ as compared with control group, (b) $p < 0.01$ as compared with PBDE 209 (10 mg/kg) group. (c) $p < 0.01$ as compared with PBDE 209 (500 mg/kg) group, (d) $p < 0.01$ as compared with PBDE 209 (1500 mg/kg) group.