

The Michigan PBB Cohort 20 Years Later: Endocrine Disruption?

Project Scope

A number of observations suggest that environmental contaminants are causing endocrine disruption in humans and wildlife resulting in a variety of health outcomes. The suspect chemicals include halogenated organics, which appear to have endocrine disrupting activity, are widespread in the environment, are resistant to degradation, and accumulate in fatty tissue. Examples of these compounds include DDT (and its degradation product DDE), polybrominated biphenyls (PBBs), polychlorinated biphenyls (PCBs), polychlorinated dibenzo[*p*]dioxins (PCDDs), and polychlorinated dibenzofurans (PCDFs). Some of these compounds (DDT, DDE, some PCB congeners) are estrogenic *in vitro*, and others (other PCB congeners, TCDD) have been found to be anti-estrogenic. DDT, PCBs, PBBs, PCDDs and PCDFs are known to induce cytochrome P450 isoenzymes, possibly altering metabolism of endogenous steroid hormones. Correlations have been previously noted between the introduction of these chemicals into the general environment and rising incidence of breast, prostate and testicular cancer, cryptorchidism, ectopic pregnancies, and early endometriosis. Similarly, the presence of these environmental estrogens and a decline in sperm has been suggested.

In the early 1970s, some Michigan residents were exposed to brominated flame retardants when the flame retardant PBB was mistakenly mixed with cattle feed instead of a nutritional supplement manufactured at the same facility. Several thousand farm families and nearby residents consumed PBB-contaminated beef and dairy products. In 1976-77, the Michigan Department of Community Health (MDCH) identified 4000 exposed individuals and obtained baseline health information and serum samples.

The resulting registry included 1900 adult women and their daughters. MDCH has followed this cohort for more than 20 years, updating their files and enrolling children born to exposed mothers. This cohort, by virtue of its well-characterized exposure to PBBs and the long follow-up period, can provide valuable data on the potential endocrine disrupting effects of PBBs. The overall objective of this research project was to investigate (1) the occurrence of selected reproductive health outcomes in PBB-exposed women and their children; and (2) if there is evidence of endocrine disruption in this population associated with PBB exposure.

Grant Title and Principal Investigator

The Michigan PBB Cohort 20 Years Later: Endocrine Disruption?

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Key Findings

- This grant supported a major epidemiological investigation of a unique cohort of reproductive age women and their daughters who were exposed to polybrominated biphenyls (PBBs).
- Breastfed daughters exposed to high levels of PBB *in utero* had an earlier age at menarche than breastfed girls exposed to lower levels of PBB in utero or girls who were not breastfed, even after adjustment for potential confounders.
- Women with high exposures to PBB in serum and who had recently lost weight had shorter menstrual cycles and longer bleed lengths than women whose exposure levels were undetectable in serum and experienced a recent weight loss.
- PBB or PCB exposure, measured upon enrollment, is not associated with time to menopause, uterine fibroids, risk of endometriosis, or self-reported physician-diagnosed benign breast disease among women followed for over 20 years.
- The associations observed here support the hypothesis that pubertal events and menstrual cycle characteristics may be affected by pre- and postnatal exposure to organohalogenes.

Research under this grant resulted in eight peer-reviewed publications.

The study involved the administration of a telephone interview to the approximately 1900 women to ascertain the incidence of endocrine-sensitive endpoints such as endometriosis, benign breast disease, abnormal Pap smears, uterine fibroids, thyroid dysfunction, and infertility. Time to pregnancy for each pregnancy following exposure was ascertained. Medical records were obtained and reviewed to verify each of the clinical conditions. Developmental milestones were also tracked in daughters of the exposed women.

Relevance to ORD's Multi-Year Research Plan

This project contributes directly to the Long-Term Goal 2 (LTG-2) of ORD's MYP by providing a better understanding of the extent of the impact of EDCs on human health.

A large cohort of women, who were exposed to polybrominated biphenyls (PBBs) in the early 1970s through PBB-contaminated beef and dairy products, were followed and evaluated for a number of endocrine-sensitive endpoints including pubertal development, menstrual function, pregnancy outcomes, endometriosis, and benign breast disease. Individual exposure levels were determined by measurement of PBBs in serum at enrollment into the cohort, as well as a number of subsequent time points. The most important positive finding of this research was that the onset of puberty in girls occurred earlier among those exposed to PBBs *in utero* and through breastfeeding. This result has opened up a new area of research into the contribution of environmental exposures to pubertal development which has consequences for the risk of breast cancer. Most studies of environmental contributions to breast cancer have focused on adult exposure and have been negative. However, if environmental exposures can accelerate pubertal development, this would increase the risks of early sexual activity, adverse pregnancy outcomes, and an increased risk of breast cancer.

Project Results

Activities completed under this grant included: (1) completion of the telephone interviews; (2) data cleaning activities and data set construction; (3) implementation of the medical verification process; (4) development of a mathematical model of serum PBB decay; and (5) analyses of relationships between exposure and health outcomes. The field phase of the study was completed in March 1998. A total of 1,185 telephone interviews were completed. The overall participation rate was 85 percent. A database was generated by the Michigan Public Health Institute and provided to the investigators for cleaning and analyses and to the MDCH for updating of the registry. Archived data collected by the MDCH were also used to clarify responses and provide missing data. Exposure data were obtained from the MDCH for PBB, polychlorinated biphenyls (PCB), and pesticides. Diagnostic verification of reported outcomes that had not been confirmed by MDCH was completed for all clinical conditions reported by participants. Medical records were obtained for diagnoses of thyroid disorders, endometriosis, abnormal pap smears, benign breast disease, adverse pregnancy outcomes, infertility, menstrual cycle abnormalities, and uterine fibroids.

Age at Menarche and Tanner Stage in Girls Exposed *in Utero* and Postnatally to PBB

The investigators assessed pubertal development in females 5-24 years of age (N=327) who were exposed to PBB *in utero* and, in many cases, through breastfeeding. They estimated *in utero* PBB exposure using maternal serum PBB measurements taken after exposure (1976–1979) and extrapolated to time of pregnancy using a model of PBB decay. They found that breastfed girls exposed to high levels of PBB *in utero* (≥ 7 parts per billion) had an earlier age at menarche (mean age = 11.6 years) than breastfed girls exposed to lower levels of PBB *in utero* (mean age = 12.2 – 12.6 years), or girls who were not breastfed (mean age = 12.7 years, Figure 1.) This association persisted after adjustment for potential confounders. Perinatal PBB exposure was associated with earlier pubic hair stage in breastfed girls, but

little association was found with breast development. The associations observed here lend support to the hypothesis that pubertal events may be affected by pre- and postnatal exposure to organohalogens.

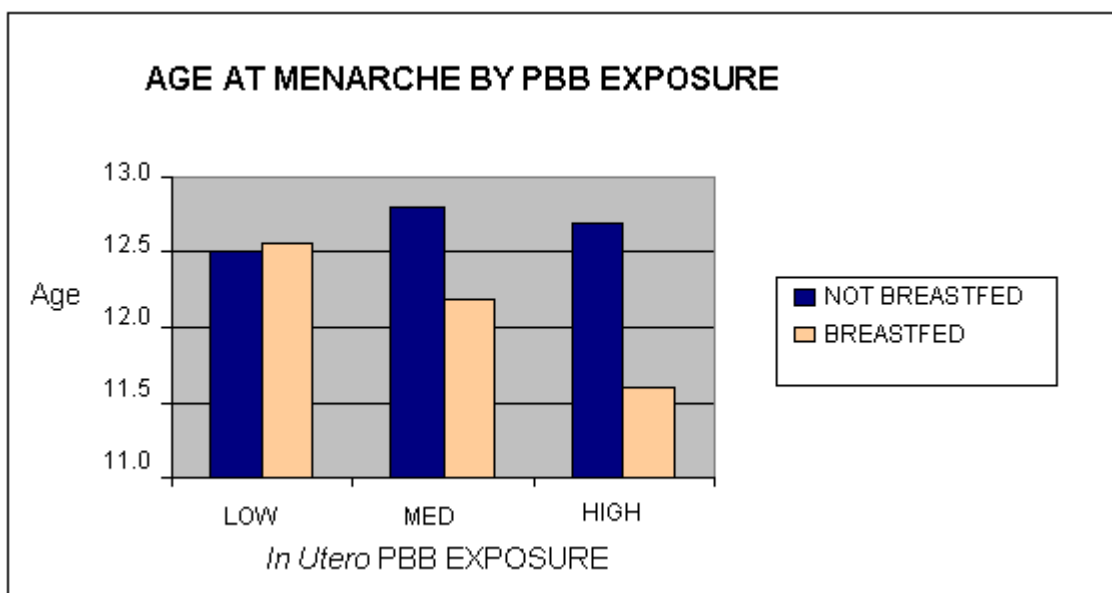


Figure 1. Average age at menarche of girls exposed to PBB *in utero* and through breastfeeding. *In utero* PBB exposure was estimated using maternal serum PBB measurements taken after exposure and extrapolated to time of pregnancy using a model of PBB decay.

Age at Menopause in Relation to PBBs, PCBs, and Smoking

Because halogenated biphenyl exposure is suspected to disrupt endocrine function, investigators assessed time to menopause in women aged 18 years and older who were orally exposed to PBBs and PCBs (n = 874). To define menopausal status, women were interviewed in 1997, and asked whether they had had any menstrual periods in the previous year, why their menstrual periods had stopped (for example, after hysterectomy), and age at their last menstrual period. Serum PBB and PCB measurements taken at enrollment (1976-1978) were used as estimate halogenated biphenyl exposure. Women whose menopause occurred before their PBB exposures were excluded. The investigators found no association between PBB or PCB exposure and age of menopause, although women who were current smokers had earlier menopause on average than never smokers, and menopause was shortest among women who were heavy current or past smokers.

PBB Exposure and Menstrual Cycle Characteristics

Adult women with at least one serum PBB measurement were eligible for the analyses of menstrual cycle length. Women using oral contraceptives or other hormonal medications were excluded, leaving 335 women for analyses. Study investigators estimated serum PBB level at the time of the interview using the initial PBB serum measurements and the prediction equation developed previously. The relationships between PBB exposure (initial and estimated) and average menstrual cycle length, and average menstrual cycle bleed length (in the past year) were evaluated by multivariate linear regression, controlling for confounders. PBB exposure and recent weight loss were found to jointly affect menstrual cycles. There was a negative association between menstrual cycle length and bleed length among women who had experienced a weight loss of more than 10 percent in the past year. Women in the highest decile of both PBB exposure and weight loss experienced menstrual cycles that were 3 to 5 days shorter than women with lower exposure or women who had not experienced weight loss. Bleed length was significantly longer (0.87 days) among women with weight loss in the highest exposure category compared to women with lower exposure or without weight loss.

Uterine Fibroids

The relationship between serum PBB measurements taken at enrollment (1976-1979) and time of diagnosis of uterine fibroids of 942 participants was estimated using multivariate models. No association was found between serum PBB level and time to diagnosis of uterine fibroids. Relative risks for women with high and moderate PBB exposure were not significantly different from women with little or no exposure.

Benign Breast Disease

The relation between serum polybrominated biphenyl (PBB) levels and the risk of benign breast disease in the Michigan cohort was examined, using extended Cox models to generate adjusted hazard ratios. Variables in the models included polychlorinated biphenyls (PCBs) and risk factors for benign breast disease reported in the literature. Two hundred fourteen (23%) of 951 women reported benign breast disease diagnosed by a physician. Neither PBB nor PCB exposure levels were found to be associated with benign breast disease. Age, smoking, and annual number of health-care provider visits were significantly associated with benign breast disease. A relationship between breast disease and smoking had not previously been observed.

Risk of Endometriosis

Statistical models were constructed to estimate the risk of endometriosis in the Michigan cohort adjusting for serum PCB level and risk factors for endometriosis reported in the literature. Ninety (9 percent) out of 970 women reported endometriosis diagnosed by a physician. Neither moderate nor high PBB exposures were found to be significantly associated with increased risk of endometriosis. A significantly increased risk of endometriosis was found among women exposed to moderate PCB levels (> 5-11 ppb), (HR = 1.85, 95 percent CI: 1.17, 2.94) but not high PCB levels (> 11 ppb), (HR = 1.29, 95 percent CI: 0.54, 3.11) compared to women with low PCB exposure (\leq 5 ppb). The reason for the observed pattern of endometriosis risk with varying PCB exposures was not explained.

Analyses of data for the following health outcomes were ongoing at the end of the funding period:

Time to Pregnancy

A total of 478 women from the Michigan PBB cohort became pregnant after exposure to PBBs. Crude analyses using Kaplan-Meier survival curves revealed a slightly shorter time to pregnancy for the highest exposure group (\geq 11 ppb) compared to women with exposures below the level of detection (\leq 1 ppb). Detailed statistical analyses of this data were being undertaken.

Abnormal Pap Smears

931 women were included in an analysis of the frequency of abnormal Pap smears and the nature of the abnormality. For all consenting participants who self-reported on a telephone interview that they have had an abnormal Pap smear, medical records were requested for the approximate time period they indicated the abnormal Pap had occurred (for example, their self-reported age at abnormal Pap, with their year of birth revealed the approximate year of the abnormal Pap). Medical records were independently reviewed by two of the investigators and results were compared. Findings of inflammation, atypia, and dysplasia were confirmed.

Risk of Osteoporosis and Bone Fractures

Women 50 years old and older in 1997 were included in an analysis of osteoporosis and bone fracture incidence. Multivariate survival analyses were performed to assess the relationship between incidence of osteoporosis and PBB exposure. Preliminary findings suggested reduced incidence of osteoporosis in PBB-exposed women, but the numbers of subjects with osteoporosis or bone fractures were small, and additional analyses were being performed combining these groups as indicators of bone density.

Summary of Major Findings

This grant supported a major epidemiological evaluation of a unique cohort of PBB-exposed reproductive age women and their daughters. The most important positive finding was that maternal PBB exposure and/or exposure through breastfeeding seemed to cause earlier onset of puberty in their daughters. This result has opened up a new area of research into the contribution of environmental exposures to pubertal development with important consequences for breast cancer.

Investigators

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For More Information

NCER Project Abstract and Reports:

http://cfpub2.epa.gov/ncer_abstracts/index.cfm/fuseaction/display.abstractDetail/abstract/137/report/0