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Polybrominated diphenyl ethers and incident pregnancy loss: The LIFE Study

Giehae Choi[a,b], **Yu-Bo Wang**[c], **Rajeshwari Sundaram**[c], **Zhen Chen**[c], **Dana Boyd Barr**[d], **Germaine M. Buck Louis**[b,e], and **Melissa M. Smarr**[b,d,*]

[a]Department of Epidemiology, University of North Carolina at Chapel Hill, Chapel Hill, North Carolina, USA

[b]Office of Director, and Bioinformatics Branch, Division of Intramural Population Health Research, *Eunice Kennedy Shriver* National Institute of Child Health and Human Development, National Institutes of Health, Bethesda, Maryland, USA

[c]Biostatistics and Bioinformatics Branch, Division of Intramural Population Health Research, *Eunice Kennedy Shriver* National Institute of Child Health and Human Development, National Institutes of Health, Bethesda, Maryland, USA

[d]Department of Environmental Health, Rollins School of Public Health, Emory University, Atlanta, Georgia, USA

[e]Dean's Office, College of Health and Human Services, George Mason University.

Abstract

Background—Polybrominated diphenyl ethers (PBDEs) have not been studied in relation to incident pregnancy loss in human populations, despite their ubiquitous exposure and purported reproductive toxicity.

Objectives—To investigate the association between preconception serum PBDE concentrations and incident pregnancy loss.

Methods—A preconception cohort of 501 couples was followed while trying to become pregnant, and for whom serum concentrations of 10 PBDE congeners were measured using gas chromatography-high resolution mass spectrometry. Pregnancy was prospectively identified as a positive home pregnancy test on the day of expected menstruation. Incident pregnancy loss was defined for 344 singleton pregnancies as a conversion to a negative home pregnancy test, menses, or clinical diagnosis depending upon gestational age. Cox proportional hazards models were used to estimate the hazard ratios (HRs) and 95% confidence intervals (CIs) for individual and summed PBDEs and incident pregnancy loss, adjusting for relevant covariates and male partners'

[*]**Corresponding author.** Melissa M. Smarr, Department of Environmental Health, Rollins School of Public Health, Emory University, Atlanta, Georgia, 30322. melissa.smarr@emory.edu.

Declarations of Conflict of Interests

None

information. In sensitivity analyses, inverse probability weighting was used to account for couples not becoming pregnant and, thereby, not at risk for loss.

Results—The incidence of prospectively observed pregnancy loss was 28%, and the serum concentrations of PBDE congeners in females were consistently associated with a higher hazard of incident pregnancy loss. Specifically, statistically significant hazard ratios (HRs) for incident pregnancy loss were observed for lower brominated PBDE congeners: 17 (HR 1.23; CI: 1.07–1.42), 28 (HR 1.25; CI: 1.03–1.52), 66 (HR 1.23; CI: 1.07–1.42), and homolog triBDE (HR: 1.25; CI: 1.05–1.49). Findings were robust to various model specifications explored in sensitivity analyses.

Conclusions—Maternal preconception serum concentrations of specific PBDE congeners may increase the hazard of incident pregnancy.

Keywords

Incident pregnancy loss; polybrominated diphenyl ethers; Longitudinal Investigation of Fertility and the Environment Study

Introduction

Polybrominated diphenyl ethers (PBDEs) are brominated flame retardants used in consumer products such as textiles, plastics, and furniture containing polyurethane foam (Dishaw et al., 2014; Siddiqi et al., 2003). PBDEs may comprise up to 209 different congeners depending on the number and positioning of bromine (Br) atoms. Previous studies suggest the number of the Br atoms to be related to the toxicity of PBDEs and prompting their grouping by the number of Br atoms into what are known as homologs (i.e., 1–10 Br). All PBDEs bioaccumulate in humans due to their long half-lives and high affinity for fat, but especially the homologs with fewer Br atoms (e.g., tetra, penta, and hexa) (Siddiqi et al., 2003). Many PBDEs have been associated with adverse health outcomes (Siddiqi et al., 2003). Three commercial brominated diphenyl ether (BDE) mixtures exist, PentaBDE comprising mostly of homologs tetra and penta; OctaBDE comprising mostly of homologs hepta and octa; and DecaBDE comprising mostly of homolog deca. PentaBDE and OctaBDE were phased out in the U.S. in 2004, and DecaBDE in 2013 (U.S. EPA 2013). Despite such policy efforts, human exposure to PBDEs remains through diet and inhalation routes of exposure (Sjödin et al., 2004a; Parry et al., 2018). Detectable concentrations of PBDE congeners in human serum may also be the result of decomposition of higher brominated congeners into lower ones (Dishaw et al., 2014; Siddiqi et al., 2003).

PBDEs are known endocrine disruptors and neurotoxins, primarily targeting the thyroid system due to their structural similarities to endogenous thyroid hormones (Siddiqi et al., 2003; Herbstman 2014). Previous studies have reported increased risk of adverse female reproductive outcomes and fetal development in relation to increased PBDE concentrations, as suggested by their associations with altered hormone milieu (Gao et al., 2016; Vuong et al., 2015), failed embryo implantation (Johnson et al., 2012), a longer time-to-pregnancy (Buck Louis et al., 2013; Gao et al., 2016), pregnancy complications (Smarr et al., 2016), and diminished birth size (Robledo et al., 2015). However, most previous research focusing

on reproductive outcomes has relied upon cohorts that enrolled pregnant women (Harley et al., 2010; Stapleton et al., 2011; Vuong et al., 2015) and often later in pregnancy or the peak window for loss.

To our knowledge, no previous studies have evaluated serum PBDE concentrations measured before conception in relation to incident pregnancy loss despite its high ≈30% incidence (Wang et al., 2003; Wilcox et al., 1988; Zinaman et al., 1996), and implications for maternal health (Engelhard et al., 2001). We sought to assess the relation between PBDEs and incident pregnancy loss, and to assess the robustness of findings when considering both maternal and paternal preconception serum PBDE concentrations.

Methods

Study population and cohort

The Longitudinal Investigation of Fertility and the Environment (LIFE) Study recruited 501 couples discontinuing contraception to try for pregnancy between 2005 and 2009 from 16 counties in Michigan and Texas. Enrolled couples met the following inclusion criteria: females aged 18–40 and males ≥18 years; in a committed relationship; no physician diagnosis of infertility/sterility; females had menstrual cycles between 21 and 42 days consistent with use of a fertility monitor to help achieve pregnancy; no injectable hormonal contraceptives in the past year which could delay the return of ovulation; and an ability to communicate in English or Spanish (Buck Louis et al., 2011). For the present analysis, the cohort was restricted to 344 (69 %) couples who had an observed singleton pregnancy while trying to conceive. This restriction recognizes that pregnancy is a necessary but insufficient criterion for loss; hence, only pregnant women are at risk for loss.

Data collection

Home visits were conducted with couples and included a baseline interview which each partner to capture data on sociodemographic characteristics, reproductive history, and lifestyle. Standardized anthropometric assessments were performed by research assistants to estimate body mass index (BMI; weight/height^2 [kg/m^2]). Non-fasting blood specimens (≈20cc) were obtained from couples using phlebotomy equipment that was selected by the laboratory to minimize possible contamination. An established protocol was used for the quantification of all PBDEs and collected in two 10-ml red top tube sterile vacutainers, whose lot was tested for PBDE contamination using our standard method with bovine serum as a surrogate matrix, using the same analytical method used to measure PBDEs in the serum samples (Sjödin et al., 2004b). Serum samples were then stored in plastic cryovials that were similarly lot-tested for contamination, and stored at −20°C or colder until shipment to the toxicology laboratory where they were processed and aliquoted per the study protocol.

Enrolled couples were followed until delivery or up to 12 months of attempting pregnancy, whichever came first. During follow-up, couples completed daily journals about their lifestyle on a daily basis through 7 weeks post conception (Buck Louis et al., 2016). In an open-ended text filed, couples were asked to list any medication taken along with indication and amount. This field was added to monitor for certain tetracyclines that are reported by the

manufacturer to potentially give inaccurate fertility monitor prompts. Menstrual cycles were defined utilizing journals supplemented with fertility monitors as needed to define menstrual cycles distinct from episodic bleeding. Specifically, a menstrual cycle denoted the interval (in days) from the onset of bleeding with a duration of ≥2 days with increasing intensity to the onset of the next similar bleeding episode. Pregnancy was defined as a positive hCG confirmed test on the day of expected menstruation. Maternal lifestyle variables during preconception and early pregnancy were derived by averaging data obtained from female partner's daily journals, and similarly for male partners' preconception lifestyle variables.

Estimation of incident pregnancy loss

Female partners were trained by research personnel and instructed in the use of the Clearblue® Easy fertility monitor to help them time intercourse to ovulation. The accuracy of this urine-based fertility monitor relative to serum measurement of luteinizing hormone (LH) as an ovulation proxy was reported to be 99% among ovulatory cycles (Behre et al., 2000). Pregnancy was identified prospectively using Clearblue® digital home pregnancy test, where women tested on the day of expected menses. The accuracy of this test kit has been reported in detail elsewhere (Johnson et al., 2015). Incident pregnancy loss was measured as conversion to a negative hCG pregnancy test, or onset of menstruation or clinical confirmation depending on gestational dating.

Laboratory analysis

Ten PBDE congeners were quantified in both partners' serum: 2,2',4-Tribromodiphenyl ether (BDE-17), 2,4,4'-tribromodiphenyl ether (BDE-28), 2,2',4,4'-tetrabromodiphenyl ether (BDE-47), 2,3',4,4'-tetrabromodiphenyl ether (BDE-66), 2,2',3,4,4'-pentabromodiphenyl ether (BDE-85), 2,2',4,4',5-pentabromodiphenyl ether (BDE-99), 2,2',4,4',6-pentabromodiphenyl ether (BDE-100), 2,2',4,4',5,5'-hexabromodiphenyl ether (BDE-153), 2,2',4,4',5,6'-hexabromodiphenyl ether (BDE-154), and 2,2',3,4,4',5',6-heptabromodiphenyl ether (BDE-183). Serum PBDEs were measured using gas chromatography-high resolution mass spectrometry with isotope dilution calibration at the Division of Laboratory Sciences, National Center for Environmental Health, Centers for Disease Control and Prevention (Sjödin et al., 2004b). All PBDE congeners were reported as nanogram per gram of serum (ng/g). The limit of detection (LOD) was 0.003 ng/g for all congeners except for BDE-47 (LOD=0.011 ng/g) and BDE-99 (LOD=0.010 ng/g). Serum lipids were quantified via commercially available enzymatic methods (Akins et al., 1989) and reported as total serum lipids (ng/g) using established methods (Phillips et al., 1989). To avoid bias produced by substituting estimates <LOD with a constant when estimating human health outcomes (Lubin et al., 2004; Schisterman et al., 2006) or by automatic lipid adjustment (Schisterman et al., 2005), the original machine-observed values were used for analyses.

Statistical analysis

Time to incident pregnancy loss was estimated by calculating the interval (days) between estimated date of conception and incident pregnancy loss. Conception was defined as the day of 'peak fertility' as indicated by the LH monitor in light of the egg's short survival time (Weinberg and Wilcox 1995). . Couples who withdrew from the study for irrespective of

reason were censored from analysis (Buck Louis et al., 2011). As all incident pregnancy losses occurred within 22 weeks post-conception, which prompted us to truncate losses at 133 days for analysis. Details on the cumulative incidence of loss in LIFE were previously described in detail (Buck Louis et al., 2016). In brief, 1% of losses occurring in the first 10 days, increasing to 15% by 30 days and 28% within our truncated window.

The distributions of sociodemographic characteristics were compared between partners and pregnancy loss status, where statistical significance was tested using the chi-square test for categorical variables and the Wilcoxon-Mann-Whitney test for continuous variables. Differences in the PBDE congener concentrations by partners, pregnancy loss and pregnancy achievement status using Wilcoxon signed-rank test and Wilcoxon-Mann-Whitney tests. Less than 10% of PBDE congeners and covariates (i.e., serum lipids, weight, height, age, and lifestyle data) were missing in the original dataset. et al., A total of 10 multiply-imputed datasets were generated using Markov chain Monte Carlo method under the missing at random assumption (Schafer 1997) to minimize potential bias (Desai et al., 2011; White and Carlin 2010).

In addition to the individual PBDE congeners, grouping by homologs and ratios of select congeners were also explored as exposure variables in separate models. Grouping by homolog is used as a basis for policy regulations (EPA 2013). Four homologs (i.e., tri: BDE-17, −28; tetra: BDE-47, −66; penta: BDE-85, −99, −100; and hexa: BDE-153, −154) were generated by summing across the molar concentrations of each congener in the homolog group. Of these homologs, tetra and penta are listed for elimination in a Stockholm convention amendment, 2009 (Xu et al., 2013), and are major components of PentaBDE mixtures (EPA 2013). For the generation of ratio measures, the ratio of molar concentrations for BDE-153 to BDE-47, and BDE-99 to BDE-47 were calculated. Exposure pattern of PentaBDE in relation to other commercial mixture may be represented by the ratio of BDE-153 to BDE-47 (Bradman et al., 2012). Ratio of BDE-99 to BDE-47 in dust and relevant human populations have been reported in previous studies (Castorina et al., 2011).

Female partners' PBDE concentrations in relation to time to incident pregnancy loss were modeled using Cox proportional hazard modeling techniques. Covariate adjustment was aided by a directed acyclic graph, and included age (years), serum lipids (ng/g), BMI (kg/m^2), race/ethnicity (non-Hispanic Whites vs. others), and cigarette smoking while pregnant (average #/day) (Buck Louis et al., 2016; Buck Louis et al., 2017; Jukic et al., 2016; Louis et al., 2016; Mu et al., 2015; Peng et al., 2016; Pollack et al., 2011; Venners et al., 2005). Proportional hazards assumption was tested using Supremum test with significant level of 0.05. Given that pregnancy is a couple-dependent outcome, we additionally generated models to include the male partners' PBDE concentrations in addition to the female partners' PBDE concentrations.

As sensitivity analyses, two additional couple-based analyses were undertaken to address different formalized assumptions about male partners' concentrations (results reported in supplementary figures). Specifically, separate models of female partners' serum PBDE congeners and incident pregnancy loss included: 1) a ratio of male to female partner's concentration or 2) total serum concentrations of PBDEs for female and male partners to

assess additivity apart from collinearity. Additionally, logistic regression was conducted to estimate and assign inverse probability censoring weights (IPCW) of achieving an observed singleton pregnancy accounting for those couples not becoming pregnant (Buck Louis et al., 2017; Rotnitzky and Robins 2005) (results reported in supplementary figures). Specifically, male partners of couples not achieving pregnancy had higher BDE-183 concentrations in comparison to those achieving (0.0021 and 0.0017ng/g, respectively) (Buck Louis et al., 2013). Therefore, IPCWs were applied to account for potential selection bias considering predictors for achieving pregnancy, including male BDE-183 concentrations.

All analyses were conducted with SAS (version 9.4; SAS Institute Inc., Cary, NC).

Results

Incident pregnancy loss inclusive of two ectopic pregnancies was 28% in the study, with all losses occurring before 22 weeks post-conception. The study population comprised mostly non-Hispanic White couples (Table 1). Age, BMI, and lipid levels were higher in males compared to females, irrespective of pregnancy loss status (Table 1).

In the study population, the highest median preconception concentrations were detected for BDE-47 (females: 0.1138 ng/g; males: 0.1052 ng/g) compared with other congeners (Table 2). The median BDE −153 and −183 concentrations were higher in males (0.0524 ng/g and 0.0017 ng/g, respectively) compared with their female partners (0.0421 ng/g and 0.0014 ng/g, respectively). No statistically significant differences in concentrations of PBDE congeners were observed by incident pregnancy loss status in the study population (Table 2).

Female partners' serum concentrations for nine out of ten PBDE congeners were associated with an increased hazard of incident pregnancy loss even after adjustment for female covariates, as shown in Table 3. Specifically, the hazard ratios of three lower brominated PBDEs ranged from 1.16 to 1.19 and were statistically significant: BDE −17 (HR: 1.19, 95% CI: 1.04–1.37), BDE-28 (HR: 1.19; 95% CI: 1.05–1.36), and BDE-66 (HR:1.16; 95% CI: 1.03–1.30). This finding was corroborated in a separate model of the homolog triBDE, which comprises BDE −17 and −28, as the main exposure (HR: 1.19, 95% CI: 1.05–1.35). All PBDE classification yielded satisfied proportional hazards assumption, with the exception of a few violations observed in 99/47.

When adjusting for male partners' PBDE congeners, the association between female partners' PBDEs and incident pregnancy loss remained with some estimates increasing in magnitude. In couple based models, four additional associations were observed: BDE −47 (HR: 1.31, 95% CI: 1.00–1.71), BDE-85 (HR: 1.26, 95% CI: 1.04–1.53), BDE-99 (HR: 1.28, 95% CI: 1.02–1.61), and BDE-154 (HR: 1.22, 95% CI: 1.03–1.45). Also, of note was the increase in the hazard of incident pregnancy loss observed for two additional homologs in females: tetraBDEs (HR: 1.26, 95% CI: 1.04–1.54) and pentaBDEs (HR: 1.23, 95% CI: 1.04–1.46.).

The association between PBDEs and pregnancy loss was robust to alternative couple-based modelling approaches in sensitivity analyses. In particular, statistically significant associations remained between BDE-28 and pregnancy loss for models of 1) female

partners' concentrations adjusting for the ratio of male to female PBDE concentrations and both partners' covariates; and 2) the sum of female and male partners' PBDE concentrations adjusting for both partners' covariates (see Fig.S1.). The results from IPCW models were similar to those observed in the primary analysis (see Fig.S2.).

Discussion

In this novel assessment of women's preconception serum PBDE concentrations and incident pregnancy loss, we found most PBDE congeners to be associated with incident pregnancy loss. Specifically, serum concentrations of lower-brominated PBDE congeners (i.e., BDE-17,−28, and −66; homolog tri) were associated with modest increases (HRs ranging: 1.22 to 1.31) in the hazard of incident pregnancy loss. These findings were robust to various models of PBDE classifications as the primary exposure and other model specifications, tested in an array of sensitivity analyses. Our findings are strengthened by the preconception recruitment of couples and daily follow up allowing for the prospective capture of incident hCG pregnancy loss (Buck et al., 2003; Buck Louis et al., 2016).

The increased risk of incident pregnancy loss observed in the current study was noticeable in lower-brominated PBDE congeners and homologs. The direction of association remained when male partner's information was considered in varying modeling scenarios and with IPCW to account for couples not achieving pregnancy. Such robust findings are noteworthy, given that we found serum PBDEs in our cohort to be much lower than the national median wet-weight values of serum PBDEs (estimated by downloading the publicly available National Health and Nutrition Examination Survey [NHANES] data on PBDES in 2005–2006 and 2007–2008 for non-lipid adjusted comparisons in females aged 20–39, Table S1). Direct comparison of our findings with previous research is challenging because of the scarcity of preconception cohorts and even fewer that are couple based. This is the first study to show that elevated preconception serum PBDEs may be associated with increased hazard of incident pregnancy loss, especially giving weight to lower brominated PBDE congeners. However, a previous study in China reported an increased odds of a threatened abortion in relation to BDE-28 (Odds Ratio (OR): 1.03; 95% CI: 0.96–1.10), and statistically significant associations with BDE-85 (OR: 1.30; 95% CI: 1.03–1.62), −153 (OR: 1.04; 95% CI: 1.01–1.08), and −183 (OR: 1.03; 95% CI: 1.01–1.06) (Gao et al., 2016). Differences between study results could be due to heterogeneity in study designs (i.e., selection criteria, timing and measurement of exposures, and model specification and techniques) and definition of pregnancy loss.

Given such robust findings supporting PBDEs as risk factors for pregnancy loss in the current study, biologic plausibility of the observed associations requires discussion. One previous study (Usenko et al., 2011) reported increased mortality in zebrafish with embryonic exposure to lower brominated PBDE congeners (−28, −47, −99, and −100) but not with higher brominated congeners (−153 and −183). Furthermore, a recent review article summarized possible biological mechanisms underlying pregnancy loss and PCBs, which is structurally similar to PBDEs, as directly affecting the developing fetus or indirectly influencing the embryo *via* alteration of the endometrium of pregnancy (Krieg et al., 2016). The biological potential for direct fetal PBDE exposures is supported by its reported toxicity

in reproductive and developmental endpoints (ATSDR. 2017), and previous studies highlighting their ability to cross the placental barrier, specifically BDE-28 (Johnson et al., 2012; Zhao et al., 2013). PBDEs may also influence embryo implantation as reported in a study of 65 women undergoing *in-vitro* fertilization (Johnson et al., 2012). The potential impact of PBDEs on early pregnancy outcomes may be explained via interference with hormonal signaling pathways (e.g., altering maternal thyroid hormonal functions (Chevrier et al., 2010; Vuong et al., 2015), exhibiting estrogenic activity (Harley et al., 2010)) or increasing inflammatory reaction (Peltier et al., 2012; Yuan et al., 2017), all of which could in turn interfere with maternal-fetal interface (Mor et al., 2011).

Cautious interpretation of our findings is warranted given important limitations associated with this observational study. These considerations include attention to residual confounding, limited statistical power for congeners skewed toward lower concentrations, and imperfect outcome measurement given the inability to measure pre-implantation pregnancy losses in the general population. We also recognize that pregnancies with hCG concentrations below analytical sensitivity limits for our test kits may have been missed, though we have no empirical evidence to support a systematic association with PBDE concentrations. Another limitation is in the reliance on a single preconception serum measurement from non-fasting blood samples that were collected at the convenience of study participants. However, comparable concentrations during pregnancy are suggested by studies reporting long half-lives (3.0–11.7 years; Geyer et al., 2004) and high intra class correlations (0.87–0.99) estimated from three repeated samples from reproductive age office workers in Boston during 2010 to 2011 (Makey et al., 2014).

The robust association found in the current study despite its limitations calls for further research in PBDEs and pregnancy loss across gestation. This is important as most pregnancy losses occurred early in gestation in our cohort in keeping with the study's preconception design. To this end, we cannot speculate about the relation between PBDEs and loss across pregnancy. We await corroboration including in populations with socio-economically diverse characteristics (Zota et al., 2010) given our finding in a well-educated and high-income population, unique exposures, or other risk profiles that may further delineate underlying mechanisms. Given the many routes for human exposure (Domingo 2012; Buttke et al., 2013) coupled with the structural persistence of these compounds, additional study is warranted to help ensure the adequacy of human protection.

Conclusion

Serum PBDE concentrations in females were associated with increased hazard of incident pregnancy loss in this first prospective study with preconception enrollment and follow-up of pregnant women to delivery. Associations were predominantly observed for lower brominated PBDE congeners and were robust to multiple sensitivity analyses including additional adjustment for male partner's information. Reasons underlying this particular association are not well characterized, but the current study results may offer insight for further mechanistic and epidemiological research.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Highlights

- 9/10 serum PBDEs were associated with increased hazard of incident pregnancy loss
- Robust associations were observed for lower brominated PBDE congeners (17, 28, 66)
- Covariate adjustment for male partners strengthened some but not all associations

Table 1.

Sociodemographic and lifestyle characteristics of the study cohort, by incident pregnancy loss status (n=344)

	Loss (n=98)			No Loss (n=246)			p-value[a]	
Characteristic[c]	**Females**	**Males**	**p[b]**	**Females**	**Males**	**p[b]**	**Female**	**Male**
Age (year)	30.30±4.1 (98)	32.27±4.7 (98)	**<.0001**	29.59±3.8 (246)	31.33±4.5 (246)	**<0.01**	0.20	0.09
BMI (kg/m^2)	27.81±6.7 (98)	30.10±5.6 (97)	**<.0001**	26.60±6.7 (246)	29.60±5.5 (246)	**<0.01**	0.09	0.47
Lipids (ng/g)	597.32±93.8 (93)	723.27±226.4 (97)	**<.0001**	617.92±119.4 (239)	723.86±208.8 (240)	**<.0001**	0.28	0.68
Average cigarettes (#/day)	0.75±3.1 (98)	1.46±4.8 (97)	0.23	0.30±1.6 (245)	0.77±3.4 (233)	0.35	0.50	0.38
NHW	82 (85)	84 (87)	0.68	203 (83)	201 (82)	0.74	0.76	0.31

[a] Comparison by loss status using Wilcoxon-Mann-Whitney for continuous and χ^2 test for categorical variables.

[b] Comparison by gender using Wilcoxon-Mann-Whitney test for continuous and χ^2 test for categorical variables.

[c] Mean ± standard deviation (n) for continuous variables, n (%) for categorical variables.

Table 2.

Distributions of female and male partner's preconception PBDE congeners' concentrations (ng/g) by pregnancy loss status (loss=98; no loss=246) and pregnancy achievement (achieved pregnancy=study population=344; couples excluded from analysis[b]=157)

Partner	PBDE congener	Loss (n=98)		No loss (n=246)		Study Cohort (n=344, loss and no loss population)		Couples excluded from analysis [b] (n=157)	
		Median (Q1, Q3)	%<LOD [a]	Median (Q1, Q3)	%<LOD [a]	Median (Q1, Q3)	%<LOD [a]	Median (Q1, Q3)	%<LOD [a]
Female	17	0.0006 (0, 0.0022)	82	0.0004 (0, 0.0016)	88	0.0005 (0, 0.0018)	86	0.0005 (0, 0.0022)	80
	28	0.0076 (0.0033, 0.0162)	21	0.0069 (0.0028, 0.0135)	26	0.0071 (0.0032, 0.0144)	24	0.0074 (0.0018, 0.0169)	27
	47	0.1212 (0.0601, 0.2823)	0	0.1093 (0.0547, 0.2163)	0	0.1138 (0.0582, 0.2273)	0	0.1153 (0.0721, 0.2486)	0
	66	0.0005 (0, 0.0019)	86	0.0004 (0, 0.0015)	92	0.0005 (0, 0.0015)	90	0.0005 (0, 0.0016)	89
	85	0.002 (0.0011, 0.0061)	66	0.0019 (0.001, 0.0039)	65	0.0019 (0.0011, 0.004)	65	0.0023 (0.0011, 0.0045)	62
	99	0.0199 (0.0108, 0.0559)	0	0.0182 (0.0093, 0.0382)	3	0.0186 (0.0099, 0.0396)	2	0.0226 (0.0107, 0.0472)	1
	100	0.0261 (0.0111, 0.0573)	19	0.0214 (0.0117, 0.0427)	19	0.0228 (0.0117, 0.0459)	19	0.0239 (0.013, 0.0493)	14
	153	0.0419 (0.0186, 0.093)	0	0.0421 (0.0216, 0.0908)	0	0.0421 (0.021, 0.092)	0	0.0357 (0.0187, 0.0877)	1
	154	0.0018 (0.001, 0.0054)	67	0.0018 (0.0008, 0.0038)	66	0.0018 (0.0009, 0.004)	66	0.0021 (0.0013, 0.0046)	61
	183	0.0014 (0.0009, 0.0021)	81	0.0015 (0.0009, 0.0021)	87	0.0014 (0.0009, 0.0021)	86	0.0015 (0.001, 0.0022)	86
Male	17	0.0004 (0, 0.0017)	81	0.0002 (0, 0.0015)	84	0.0003 (0, 0.0016)	83	0.0003 (0, 0.0022)	83
	28	0.0073 (0.0015, 0.0163)	29	0.006 (0.0017, 0.0139)	31	0.0064 (0.0017, 0.0142)	30	0.0091 (0.0021, 0.0158)	28
	47	0.123 (0.0595, 0.2395)	1	0.0971 (0.051, 0.2226)	0	0.1052 (0.0526, 0.2247)	1	0.1396 (0.064, 0.2789)	1
	66	0.0006 (0, 0.0023)	80	0.0004 (0, 0.0014)	89	0.0005 (0, 0.0015)	86	0 (0, 0.0017)	85
	85	0.0021 (0.0009, 0.0042)	65	0.0021 (0.001, 0.004)	66	0.0021 (0.001, 0.004)	65	0.0027 (0.0011, 0.0045)	55
	99	0.0205 (0.0107, 0.0465)	1	0.0183 (0.0097, 0.0401)	0	0.0191 (0.0099, 0.0416)	1	0.025 (0.0122, 0.0552)	1
	100	0.0256 (0.0119, 0.0604)	22	0.0207 (0.0112, 0.048)	24	0.0219 (0.0113, 0.0501)	23	0.0294 (0.0137, 0.0627)	17
	153	0.0648 (0.0234, 0.1735)	0	0.0504 (0.0268, 0.1503)	0	0.0524 (0.0265, 0.1526)	0	0.0631 (0.0271, 0.1581)	0
	154	0.0021 (0.0008, 0.0055)	58	0.0019 (0.0008, 0.0043)	65	0.002 (0.0008, 0.0045)	63	0.0025 (0.001, 0.0048)	58
	183	0.0018 (0.001, 0.0026)	80	0.0017 (0.0011, 0.0027)	78	0.0017 (0.001, 0.0027)	78	0.0021 (0.0014, 0.0037)	64

[a] The limit of detection (LOD) was 0.003 ng/g for all PBDE congeners except BDE-47 (LOD=0.011 ng/g) and BDE-99 (LOD=0.010 ng/g).

[b] This group includes couples who were censored, had multiple gestations, or had no observed pregnancy

Table 3

Female partners' PBDE concentrations and time to incident pregnancy loss, expressed in hazard ratios (HRs) per 1 unit change in PBDE classification.

PBDE classification[a]	Female Model		Couple-based model	
	Unadjusted HR (95% CI)	Adjusted HR (95% CI) [b]	Unadjusted HR (95% CI) [c]	Adjusted HR (95% CI)[d]
17	**1.17 (1.02, 1.33)**	**1.19 (1.04, 1.37)**	**1.16 (1.01, 1.34)**	**1.23 (1.07, 1.42)**
28	**1.15 (1.01, 1.31)**	**1.19 (1.05, 1.36)**	1.14 (0.94, 1.37)	**1.25 (1.03, 1.52)**
47	1.13 (0.96, 1.33)	1.14 (0.96, 1.36)	1.15 (0.89, 1.48)	1.31 (1.00, 1.71)
66	**1.14 (1.02, 1.26)**	**1.16 (1.03, 1.30)**	1.13 (0.99, 1.30)	**1.23 (1.07, 1.42)**
85	1.11 (0.96, 1.28)	1.12 (0.97, 1.31)	1.16 (0.95, 1.41)	**1.26 (1.04, 1.53)**
99	1.12 (0.96, 1.30)	1.10 (0.93, 1.31)	1.15 (0.92, 1.44)	**1.28 (1.02, 1.61)**
100	1.10 (0.93, 1.30)	1.13 (0.95, 1.34)	1.04 (0.83, 1.31)	1.13 (0.89, 1.44)
153	0.96 (0.78, 1.19)	1.00 (0.81, 1.23)	0.92 (0.74, 1.15)	0.92 (0.73, 1.16)
154	1.12 (0.97, 1.28)	1.13 (0.97, 1.32)	1.13 (0.96, 1.34)	**1.22 (1.03, 1.45)**
183	1.07 (0.91, 1.26)	1.10 (0.93, 1.29)	1.08 (0.92, 1.27)	1.12 (0.94, 1.33)
Tri	**1.15 (1.02, 1.30)**	**1.19 (1.05, 1.35)**	1.14 (0.96, 1.35)	**1.25 (1.05, 1.49)**
Tetra	1.12 (0.98, 1.28)	1.14 (0.99, 1.31)	1.14 (0.93, 1.39)	**1.26 (1.04, 1.54)**
Penta	1.11 (0.98, 1.27)	1.12 (0.97, 1.30)	1.14 (0.96, 1.35)	**1.23 (1.04, 1.46)**
Hexa	1.03 (0.86, 1.25)	1.07 (0.88, 1.29)	0.98 (0.79, 1.21)	0.99 (0.79, 1.23)
153/47	0.78 (0.59, 1.05)	0.79 (0.59, 1.06)	0.79 (0.59, 1.05)	0.73 (0.52, 1.03)
99/47	1.04 (0.44, 2.50)	0.95 (0.39, 2.36)	1.00 (0.42, 2.39)	0.96 (0.38, 2.39)

[a] Congeners 17–183: log standardized concentrations of single congeners (ng/g); Tri: log standardized molar sum of BDE-17 and −28;

Tetra: log standardized molar sum of BDE-47 and −66;

Penta: log standardized molar sum of BDE −85, −99, and −100;

Hexa: log standardized molar sum of BDE −153, and −154;

153/47: log standardized ratio of molar sums of BDE −153 to −47;

99/47: log standardized ratio of molar sums of BDE −99 to −47.

[b] Female partners' concentrations modeled adjusting for her age, serum lipids (ng/g), BMI (kg/m^2), race/ethnicity (non-Hispanic Whites vs. others), and cigarette smoking (average #/day).

[c] Female partners' concentrations modeled adjusting for male partners' concentrations.

[d] Female partners' concentrations modeled adjusting for male partners' concentrations, her age, age difference between partners, and both partners' serum lipids (ng/g), BMI (kg/m^2), race/ethnicity (non-Hispanic Whites vs. others), and cigarette smoking (average #/day).