Determinants of Fetal Exposure to Polyfluoroalkyl Compounds in Baltimore, Maryland

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Polyfluoroalkyl compounds (PFCs), such as perfluorooctane sulfonate (PFOS) and perfluorooctanoate (PFOA), are ubiquitous, man-made chemicals. Human data suggest that *in utero* exposures to these chemicals occur and some evidence of developmental toxicity in animals exists. To assess the distribution and determinants of fetal exposure to PFCs, we analyzed cord serum samples from 299 singleton newborns delivered between 2004 and 2005 in Baltimore, MD for 10 PFCs by employing on-line solid-phase extraction coupled with reversed-phase high-performance liquid chromatography−tandem mass spectrometry. PFOS and PFOA were detected in 99 and 100% of umbilical cord sera, with geometric mean concentrations of 4.9 and 1.6 ng/mL, respectively. PFOS and PFOA concentrations were highly correlated (Pearson's $r = 0.64$ after natural log transformation, $p < 0.01$). Eight other PFCs were detected less frequently and at lower concentrations than PFOS and PFOA. Geometric mean concentrations of PFOS for Asians (6.0 ng/mL) and Blacks (5.1 ng/mL) were higher than those for Whites (4.2 ng/mL), while PFOA levels were more evenly distributed by race. Other maternal demographic and socioeconomic characteristics, including age, education, marital status, and living in the city limits were not significantly associated with cord concentrations. Our findings suggest that *in utero* exposure to PFOS and PFOA is ubiquitous in a population of babies born in Baltimore, MD.

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Introduction

Polyfluoroalkyl compounds (PFCs) comprise a class of man-made, fluorinated organic compounds that have been used in a variety of consumer and industrial applications for more than 50 years. These applications include protective coatings for food-contact packaging, textile, carpets, and leather, processing aids in the production of fluoropolymers, commercial and industrial surfactants, and insecticides (*1*, *2*). Only recently have reports documented widespread exposure in wildlife and humans (*3*−*6*). In 2000, the major manufacturer of perfluorooctane sulfonate (PFOS) announced a voluntary phase-out of this product (*7*). Early in 2006 the major manufacturer of perfluorooctanoate (PFOA) reported having achieved a voluntary reduction in PFOA emissions by the end of 2005 as well as a commitment for a total reduction in emissions of 98% by 2007 (*8*).

PFOS has been identified as a hepatic peroxisome proliferator that targets the liver and disrupts lipid metabolism in some animal species (*9*). Toxicity studies in animals have shown marked reductions in serum cholesterol and/or triglycerides (*10*−*12*) and endocrine (*10*, *12*−*15*), developmental, and reproductive effects (*12*, *14*−*16*). PFOS has been found to be tumorigenic and carcinogenic in rats (*17*). PFOA is also hepatotoxic (*18*), a peroxisome proliferator (*19*), and disrupts lipid metabolism in some species (*11*). PFOA has been shown to be tumorigenic in rats (*18*, *20*), and some suspect that it may be a human carcinogen (*21*). In rats and mice, PFOA has shown the potential for developmental toxicity (*22*−*24*). However, it should be noted that serum concentrations associated with toxicity in animal studies are orders of magnitude higher than those reported in humans, even those exposed occupationally.

PFCs are highly stable in the environment and the half-life in humans has been estimated at 5.4 years for PFOS and 3.8 years for PFOA (*25*). Many PFCs are surfactants; rather than accumulating in lipids like traditional persistent organic pollutants, they are bound to proteins in the liver, serum, and other tissues (*26*−*28*). PFOS and PFOA have been detected consistently in human biomonitoring studies in the United States (*5*, *29*−*31*) and many other countries (*3*), while other PFCs are not consistently found.

Despite the growing body of evidence suggesting widespread human exposure, little is known about the presence of PFCs *in utero*. A study of 15 maternal−fetal pairs in Japan confirmed that PFOS could cross the placental barrier in humans, albeit incompletely (*32*). Other small studies in Germany and Northern Canada documented detectable levels of PFOS and PFOA in cord blood samples (*33*, *34*). The aims of the current study were to characterize the distribution of serum concentrations of PFCs and to identify demographic and socioeconomic factors associated with *in utero* exposure to these chemicals among a population of babies born in Baltimore, MD from November 2004 through March 2005.

Materials and Methods

Subjects. We conducted a cross-sectional study (the Baltimore THREE Study) of newborn deliveries at the Johns Hopkins Hospital in Baltimore, MD. This study received approval from the Johns Hopkins Medicine Institutional Review Board and was determined to be exempt from the Health Insurance Portability and Accountability Act. The study required the collection only of specimens that otherwise would have been discarded and information from medical records that were available to hospital personnel. Thus, there was no requirement for informed consent due to the

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anonymization of all samples and data. Members of a community advisory committee, who were selected for their specific knowledge and expertise, and their focus on important child health concerns in Maryland, had the opportunity to learn about and comment on this study before it was conducted. Between November 26, 2004 and March 16, 2005 all singleton, live birth deliveries occurring in the Labor and Delivery Suite at the hospital were eligible for participation in the study.

Over the course of the study period, 609 live births occurred at the hospital, of which 597 were singleton births. We obtained cord blood specimens from 341 of these. We conducted a brief survey of hospital personnel to understand the major reasons for missed specimen collection. The most common explanations for missed collection included: complications during delivery, premature birth and/or small size of the infant resulting in small quantity of available cord blood, and logistical factors such as understaffing. The babies who were not included had somewhat lower gestational ages and birth weights. Forty-two of the 341 specimens collected had insufficient volume for laboratory analyses of PFCs and were excluded, leaving a total of 299 in this study. Factors associated with lower blood volumes collected were: preterm birth, low birth weight, being first born, and younger age of mother.

Cord blood samples were collected by hospital personnel immediately following delivery from the umbilical cord vein (*35*). After delivery, a section of the cord was cleaned with an alcohol wipe and blood was drawn using a sterile 60-mL Becton Dickinson (BD) syringe with an 18-gauge safety needle. A BD Vacutainer Blood Transfer Device was then attached to the syringe and up to five 10 mL glass BD vacutainers were filled. Cord blood specimens were stored in Labor and Delivery refrigerators and, within 3 h, transported to a laboratory at the Johns Hopkins Bloomberg School of Public Health for processing. Blood specimens were centrifuged at 1000*g* for 15 min. Serum was aliquotted into prescreened 2 mL polypropylene cryovials and stored at −80 °C. The prescreened containers were previously shown to be free of PFC contaminants. Frozen specimens were transferred on dry ice to the Centers for Disease Control and Prevention (CDC) for analyses.

Medical Records. Two study investigators concurrently abstracted maternal and infant characteristics from clinical databases maintained by the hospital. A random 10% sample was verified by two other investigators. Additional information was obtained from forms filled out by the nursing staff at the time of delivery. These data were collected to examine factors that may be associated with *in utero* exposure to PFCs, such as maternal birth cohort, social class, and past pregnancies. Age, race, education, marital status, and parity were based on self-report. Insurance type was recoded from the medical record as “Private” or “Public Assistance.” Body mass index (BMI) was calculated from reported pre-pregnancy weight and height. Gestational age was based on the “best obstetric estimate” and categorized as term (≥37 full weeks) or preterm (<37 full weeks). Infant sex was abstracted from medical records. Maternal smoking status during pregnancy was defined using the maternal medical record and cord serum cotinine concentrations. Cotinine concentrations of 1−10 ng/mL were categorized as passive smoking exposure and concentrations above 10 ng/mL as active smoking exposure (*36*). If the clinical record indicated that the mother smoked during pregnancy, she was considered an active smoker regardless of the cotinine concentration in cord blood. The mother’s home address was geocoded by Geolytics, Inc. Residence inside the city limits was defined using Federal information processing standards code 24510.

Laboratory Analysis. Cord serum samples were analyzed for 10 PFCs by on-line solid-phase extraction (SPE) coupled with reversed-phase high-performance liquid chromatography (HPLC)−tandem mass spectrometry (MS/MS). The method has been described in detail by Kuklenyik et al. (*37*). This method, used to measure PFCs in large-scale surveys, including the National Health and Nutrition Examination Survey (NHANES), has excellent recovery, precision, and reliability for the detection of PFCs in human serum (*37*). Briefly, without protein precipitation, one aliquot of 100 μL of serum was injected into a commercial column switching system allowing for concentration of the analytes on a SPE column. This column was placed automatically in front of an analytical column for chromatographic separation of the analytes. Detection and quantification were done using negative-ion TurboIonSpray ionization, a variant of electrospray ionization, tandem mass spectrometry. The limits of detection (LODs) were in the low ng/mL range for the following PFCs: perfluorooctane sulfonamide (PFOSA), 2-(*N*-ethyl-perfluorooctane sulfonamido) acetate, 2-(*N*-methyl-perfluorooctane sulfonamido) acetate (Me-PFOSA-AcOH), perfluorobutane sulfonate, PFOS, perfluoroheptanoate, PFOA, perfluorodecanoate (PFDeA), perfluoroundecanoate (PFUA), and perfluorododecanoate. Although the analytical method allows for the quantification of perfluorohexane sulfonate (PFHxS) and perfluorononanoate (PFNA), these analytes could not be measured in the cord sera due to the presence of interferent compounds that eluted at the same retention times and shared precursor/product ion transitions of identical mass-to-charge ratios (m/z) with PFHxS and PFNA. Similarly, the precursor/product ion m/z transition normally used for the quantification of PFOS (*37*) also had an interference. Therefore, PFOS concentrations had to be calculated using another transition, one of the two normally used to confirm the presence of PFOS (*37*). The nature of these interferences is at present unknown. Analytical standards, quality control (QC), and reagent blank samples were included in each analytical batch along with the unknown samples. QC samples were evaluated according to standard statistical probability rules.

Serum cotinine was measured by the CDC using a method described by Bernert et al. (*38*). It employs HPLC coupled with atmospheric pressure chemical ionization MS/MS to measure serum cotinine with high accuracy and sensitivity (LOD = 0.015 ng/mL). This method has been used to assess exposure to environmental tobacco smoke in NHANES and other large-scale surveys.

Statistical Analysis. We used descriptive statistics to describe cord serum PFC concentrations. Because PFC concentrations were skewed to the right, analyses utilized natural log-transformed concentrations. We used Pearson’s correlation to test for linear co-occurrence of PFCs and linear regression to describe univariate relationships between predictors and PFC concentrations. The possibility of non-linear relationships was explored using restricted cubic spline models.

We used linear regression to estimate the ratio of geometric mean concentrations (and 95% confidence intervals [95% CI]) among categories of maternal characteristics. Under the linear regression model, the expectation (or average) of the natural log PFC concentration is described as follows:

$$E(\ln PFC) = \beta_0 + \beta_1 x_1 + \epsilon$$

where β_0 is the intercept, ϵ is the normally distributed error term, x_1 is a maternal or infant predictor, and β_1 is the regression coefficient, which is equal to

$$\beta_1 = E(\ln PFC)_{x_1=1} - E(\ln PFC)_{x_1=0}$$

TABLE 1. Perfluorinated Chemicals (PFCs) Measured in Cord Blood Serum and Reported in Units of ng/mL (n = 299) from the Baltimore THREE Study, 2004–2005

compound[a]	limit of detection (LOD)	% above LOD	geometric mean[b] (range)
PFOSA	0.05	26	<LOD (ND–0.8)
Et-PFOSA-AcOH	0.2	1	<LOD (ND–0.5)
Me-PFOSA-AcOH	0.2	40	<LOD (ND–1.8)
PFBuS	0.1	3	<LOD (ND–0.2)
PFOS	0.2	99	4.9 (ND–34.8)
PFHpA	0.4	2	<LOD (ND–2.6)
PFOA	0.1–0.2	100	1.6 (0.3–7.1)
PFDeA	0.2	24	<LOD (ND–1.1)
PFUA	0.2	34	<LOD (ND–1.9)
PFDoA	0.2	5	<LOD (ND–1.7)

[a] PFOSA = perfluorooctane sulfonamide; Et-PFOSA-AcOH = 2-(*N*-ethyl-perfluorooctane sulfonamido) acetate; Me-PFOSA-AcOH = 2-(*N*-methyl-perfluorooctane sulfonamido) acetate; PFBuS = perfluorobutane sulfonate; PFOS = perfluorooctane sulfonate; PFHpA = perfluoroheptanoate; PFOA = perfluorooctanoate; PFDeA = perfluorodecanoate; PFUA = perfluoroundecanoate; PFDoA = perfluorododecanoate. [b] Non-detects (ND) are computed as the LOD/$\sqrt{2}$. Geometric mean is listed as <LOD if ≥60% of observations are <LOD.

After exponentiating the coefficient, the equation reduces to

$$e^{\beta_1} = \frac{e^{E(\ln PFC_{x1=1})}}{e^{E(\ln PFC_{x1=0})}} = \frac{GM(PFC_{x_1=1})}{GM(PFC_{x_1=0})}$$

The exponentiated coefficient can be interpreted as the ratio of geometric mean concentrations (GM) comparing one stratum of the categorical predictor, x_1, to another. The 95% CI on this ratio can be estimated similarly, by exponentiating the confidence intervals of the coefficient.

We used multivariate linear regression to compare geometric mean concentrations, after adjusting for other covariates. For all models, regression diagnostics were conducted to assess fit and the presence of heteroskedasticity. Concentrations below the LOD (<LOD) were imputed as the LOD divided by the square root of two (*39*). Statistical analyses were performed using STATA version 8.0 (StataCorp, College Station, TX).

Results

Table 1 summarizes the occurrence and concentration of PFCs detected in umbilical cord blood serum. PFOA was detected in all samples and PFOS was detected in all but two samples, with corresponding geometric means of 1.6 ng/mL for PFOA (range 0.3–7.1 ng/mL) and 4.9 ng/mL for PFOS (range <LOD–34.8 ng/mL). The 95th percentile concentration was 3.4 ng/mL for PFOA and 12.4 ng/mL for PFOS. Four other PFCs were detected in at least 20% of samples (PFOSA, Me-PFOSA-AcOH, PFDeA, PFUA). PFOS and PFOA made up most of the total concentration of the PFCs measured in these specimens. Because of the observed low detection frequency and concentrations of eight of the ten analytes, further analyses of determinants of exposure were conducted only for PFOS and PFOA.

As expected, concentrations of both PFOS and PFOA were right skewed and became more Gaussian after natural log-transformation. Cord concentrations of PFOS and PFOA were highly correlated with one another (Figure 1; Pearson's r = 0.64, $p < 0.01$).

In Table 2, the geometric mean PFOS and PFOA concentrations are shown by maternal and infant characteristics, along with the multivariate adjusted ratio of geometric means and 95% CIs. The geometric mean concentrations of PFOS for Asians (6.0 ng/mL) and Blacks (5.1 ng/mL) were higher than those for Whites (4.2 ng/mL), while PFOA levels were more evenly distributed by race. Male babies had lower geometric mean concentrations than female babies for both compounds (PFOS, $p = 0.07$; PFOA, $p < 0.01$). Obese (BMI ≥30 kg/m^2) and underweight (BMI <18.5 kg/m^2) women had babies with slightly higher geometric mean concentrations compared with normal weight (BMI 18.5–24.9 kg/m^2) women, although only statistically significant for PFOA among obese women ($p = 0.03$). Evidence of a nonlinear relationship with BMI was confirmed using restricted cubic spline models (data not shown). Multiparous births had slightly lower PFOS and PFOA cord concentrations than primiparous births, and preterm births (<37 completed weeks of gestation) had lower concentrations than term births, although the difference was statistically significant only for PFOA and parity ($p = 0.02$). There were no other significant predictors of cord concentrations among the remaining covariates, which included age, education, insurance type, marital status, smoking status, and living inside the city limits. When examining covariates as continuous measures, no significant linear trends were observed between PFOS or PFOA and maternal age, gestational age, or cord cotinine concentration (data not shown).

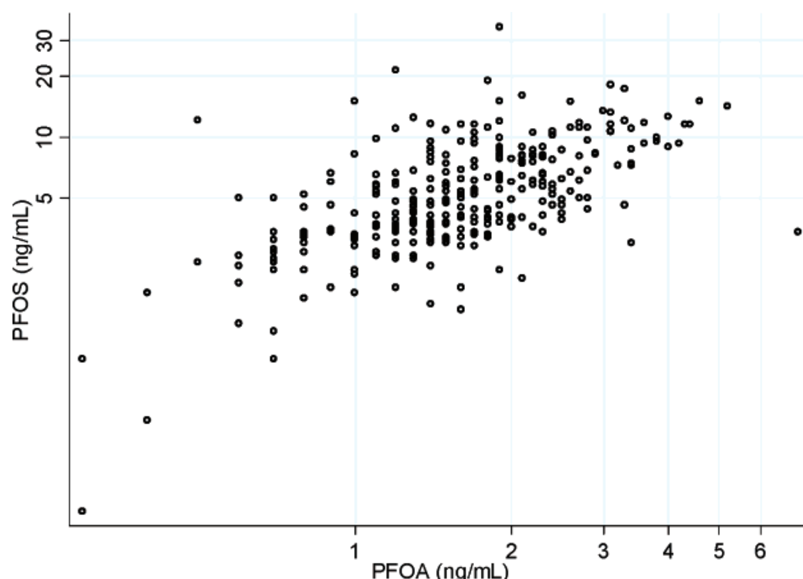

FIGURE 1. Correlation between log perfluorooctane sulfonate (PFOS) and log perfluorooctanoate (PFOA) concentrations in cord blood serum (n = 299). Pearson's r = 0.64; $p < 0.01$. Baltimore THREE Study, 2004–2005.

TABLE 2. Adjusted Ratios (and 95% Confidence Intervals) of Geometric Mean Perfluorooctane Sulfonate (PFOS) and Perfluorooctanoate (PFOA) Concentrations in Cord Blood Serum by Maternal and Infant Characteristics from the Baltimore THREE Study, 2004−2005

characteristic	*N*	PFOS		PFOA	
		GM[a] [ng/mL]	GM ratio[b]	GM[a] [ng/mL]	GM ratio[b]
maternal age					
<18 years	25	4.9 (4.0−6.2)	1.02 (0.73−1.42)	1.5 (1.2−1.8)	0.98 (0.77−1.24)
18−35 years	250	4.9 (4.5−5.4)	1.00	1.6 (1.5−1.7)	1.00
>35 years	24	5.0 (3.8−6.5)	1.13 (0.84−1.53)	1.5 (1.2−1.9)	1.07 (0.86−1.33)
maternal race					
White	64	4.2 (3.5−5.0)	1.00	1.5 (1.3−1.7)	1.00
Asian	25	6.0 (3.8−9.4)	*1.43 (1.02−2.01)*	1.5 (1.1−2.1)	1.06 (0.83−1.35)
Black	210	5.1 (4.7−5.5)	1.28 (0.98−1.68)	1.6 (1.5−1.7)	1.12 (0.92−1.36)
maternal education					
<HS diploma	87	4.8 (4.3−5.4)	1.00	1.5 (1.4−1.7)	1.00
HS diploma	97	5.1 (4.6−5.7)	1.05 (0.84−1.32)	1.7 (1.5−1.8)	1.13 (0.96−1.33)
1−4 years college	69	4.9 (4.0−5.9)	1.05 (0.79−1.41)	1.5 (1.4−1.7)	1.15 (0.93−1.42)
5+ years college	42	5.1 (3.8−6.8)	1.05 (0.70−1.57)	1.6 (1.3−1.9)	1.19 (0.89−1.60)
health insurance					
public assistance	98	4.9 (4.3−5.6)	1.00	1.6 (1.5−1.8)	1.00
private	116	5.1 (4.4−5.8)	1.10 (0.86−1.41)	1.5 (1.4−1.7)	0.95 (0.80−1.14)
marital status					
unmarried	198	5.0 (4.6−5.5)	1.00	1.6 (1.5−1.7)	1.00
married	101	4.8 (4.1−5.6)	0.90 (0.67−1.19)	1.5 (1.3−1.7)	0.99 (0.81−1.22)
maternal body mass index (kg/m^2)					
underweight (<18.5)	16	5.9 (3.7−9.2)	1.21 (0.85−1.74)	1.7 (1.3−2.2)	1.13 (0.87−1.46)
normal (18.5−24.9)	135	4.8 (4.3−5.4)	1.00	1.5 (1.3−1.6)	1.00
overweight (25−29.9)	65	4.7 (3.9−5.7)	0.96 (0.78−1.18)	1.6 (1.4−1.8)	1.04 (0.89−1.20)
obese (30+)	72	5.4 (4.8−6.1)	1.11 (0.90−1.37)	1.8 (1.6−1.9)	*1.19 (1.02−1.38)*
parity					
primiparous	125	5.2 (4.5−5.9)	1.00	1.7 (1.5−1.8)	1.00
multiparous	174	4.8 (4.4−5.2)	0.91 (0.76−1.08)	1.5 (1.4−1.6)	*0.86 (0.76−0.98)*
maternal smoking					
non/passive	243	5.1 (4.6−5.5)	1.00	1.6 (1.5−1.7)	1.00
active	56	4.5 (3.9−5.1)	0.91 (0.73−1.14)	1.6 (1.4−1.8)	1.09 (0.93−1.27)
infant sex					
female	133	5.3 (4.9−5.8)	1.00	1.8 (1.6−1.9)	1.00
male	166	4.7 (4.2−5.3)	0.86 (0.74−1.01)	1.4 (1.3−1.6)	*0.81 (0.73−0.91)*
residence within Baltimore city limits at birth					
no	92	4.8 (4.1−5.6)	1.00	1.5 (1.4−1.7)	1.00
yes	207	5.0 (4.6−5.4)	0.94 (0.75−1.17)	1.6 (1.5−1.7)	1.01 (0.86−1.19)
preterm delivery (<37 wk)					
no	260	5.0 (4.7−5.5)	1.00	1.6 (1.5−1.7)	1.00
yes	39	4.3 (3.3−5.7)	0.90 (0.70−1.14)	1.4 (1.2−1.6)	0.88 (0.74−1.05)

[a] GM = geometric mean. [b] GM Ratio = ratio of geometric means. Adjusted for all variables listed in the table. The following data were missing: 4 observations for education, 85 for insurance, and 11 for BMI. Missing data were treated as an indicator term in regression models. Italicized GM ratio indicates statistically significant ($p < 0.05$) difference from reference group.

Discussion

This study confirms earlier findings indicating that the developing fetus is exposed to persistent PFCs *in utero* and that PFOS and PFOA are the predominant polyfluoroalkyl compounds detected in cord blood (*32−34*). Cord concentrations of PFOS and PFOA were strongly correlated, despite arising from different industrial sources, implying that the pathways of human exposure to PFOS and PFOA may be similar. PFOS and PFOA, along with possible precursors (polyfluoroalkyl sulfonamides and fluorotelomer alcohols), have been identified in consumer products, house dust, water, and/or indoor air (*40−44*), and these are possible pathways of exposure. For example, Me−PFOS−AcOH is a metabolite of *N*-methyl perfluorooctane sulfonamidoethanol, which has been widely used as a stain repellent for carpets. The detection of this compound in 40% of samples may reflect exposure from contact with treated carpets (*30*). The correlation between PFOS and PFOA in blood may reflect the co-occurrence and uptake of these compounds through secondary pathways, such as food or drinking water intake. PFOS and PFOA have been detected in surface waters, suggesting that drinking water is a possible source (*44−46*). Environmental contamination of these compounds has been well-documented in regions as far away as the Arctic (*47*), raising the possibility of exposures through the food chain. PFOS (and PFOA to a lesser extent) bioconcentrate in fish and biomagnify in aquatic food chains (*4*, *45*, *46*, *48*), suggesting fish consumption as a possible pathway. In a recent study in Poland, Falandysz et al. found that individuals with high fish consumption had higher concentrations of PFOS (and PFOA to a lesser extent) in their blood relative to other groups (*49*). Further study is needed to better understand the pathways of exposure to these compounds in our population.

The analytic method used to measure human serum concentrations of PFCs has excellent precision and accuracy at concentrations of PFOS and PFOA in the range of the current study (*37*). However, in this study, the measurement error for PFOS may be greater, because the ion transition normally monitored for quantification could not be used due to an interferant. Random measurement error generally would bias bivariate associations to the null (*50*), which may contribute to the lack of observed differences in PFOS concentrations between subgroups.

Geometric mean cord PFOS and PFOA concentrations in this study were within the range of previous reports from Germany and Japan (Figure 2). Inoue et al. reported the

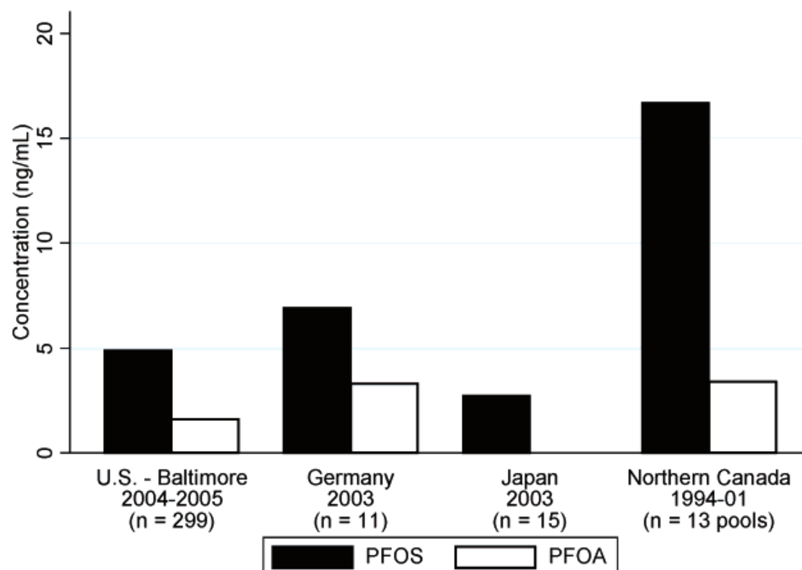

FIGURE 2. Comparison of geometric mean perfluorooctane sulfonate (PFOS) and perfluorooctanoate (PFOA) concentrations (ng/mL) in cord blood obtained for the Baltimore THREE study (2004−2005) and prior cord blood studies in Germany (*34*) (plasma), Japan (*32*) (serum), and Canada (*33*) (plasma, arithmetic mean).

presence of PFOS in all 15 cord serum samples collected in Japan, at concentrations ranging from 1.6 to 5.3 ng/mL (*32*). In the same study, PFOA was detected in only 3 maternal samples and no fetal samples (LOQ 0.5 ng/mL). In a recent German study of 11 plasma cord samples, the geometric mean PFOS and PFOA concentrations were 6.9 and 3.3 ng/mL, respectively (*34*). Finally, in a study of 13 pooled cord plasma samples in northern Canada, collected from 1994 to 2001, only arithmetic mean concentrations were reported. The means for PFOS and PFOA were 16.7 and 3.4 ng/mL, respectively (*33*), higher than arithmetic means for the present study (PFOS 6.0 ng/mL; PFOA 1.8 ng/mL).

Very few maternal or infant characteristics were predictors of cord PFC concentrations. Consistent with studies of adults, PFOS and PFOA concentrations were relatively constant across maternal age (*29*−*31*). None of the socioeconomic measures in our study (e.g., education, insurance, marital status, living in Baltimore City) were associated with PFC concentrations. Even statistically significant differences sometimes reflected minor absolute differences in dose. For example, although male babies had lower concentrations of PFOS and PFOA than females, the absolute difference in geometric means was only 0.6 ng/mL for PFOS and 0.3 ng/mL for PFOA. Overall, our results imply that cord levels are fairly uniformly distributed by maternal age and socioeconomic characteristics.

Asian and Black infants had somewhat higher PFOS concentrations than Whites. This is in contrast to an analysis of pooled serum samples from 2001−2002 NHANES, in which White females had higher levels than Black females (*5*) and to the 1999−2000 NHANES sample, in which no differences were observed between Blacks and Whites (*6*). There are several possible reasons for differences in this relationship, including random variation or ethnic differences in exposure patterns between the study populations.

The estimated lower bound of the benchmark dose associated with a 5 or 10% change in response ($BMDL_5$ or $BMDL_{10}$) can provide a useful basis for comparison. Luebker et al. estimated a $BMDL_5$ for PFOS and birth weight in rats of 0.39 mg/kg/day, equivalent to a rat fetal serum concentration of about 34 μg/mL (*15*). Butenhoff et al. reported $BMDL_{10}$s for several postnatal developmental endpoints for PFOA in rats, ranging from 22 to 44 mg/kg/day, equivalent to rat fetal serum concentrations from 29 to 59 μg/mL (*51*). By contrast, maximum concentrations in our study were 0.035 μg/mL (PFOS) and 0.007 μg/mL (PFOA). Thus, the serum concentrations of these compounds associated with developmental effects in rats are several orders of magnitude higher than what was observed here.

Our findings confirm the presence of *in utero* exposure to PFOS and PFOA, and less so, to other PFCs under study. Our data suggest that exposure is occurring among babies born in the Baltimore area, although the cord serum concentrations are lower than those reported among adults in the United States. Concentrations of PFOS and PFOA were highly correlated, possibly due to common pathways for exposure. Further, *in utero* serum concentrations of PFOS appear to be higher in Asian and Black babies when compared to White babies. What was most surprising was a lack of association between PFOS and PFOA concentrations and maternal age, socioeconomic status, and inner city residence (urban vs suburban exposures). The finding that levels were higher among obese and underweight mothers is interesting but does not have an obvious explanation. Further research to identify sources, transport, fate, and pathways of exposure to PFOS and PFOA to mothers should concentrate on general exposures, such as drinking water and commonly eaten foods. The fact that PFOA (and possibly PFOS) concentrations are slightly decreased with increased parity of mothers implies that maternal−fetal transfer may be reducing maternal stores. However, a recent study has indicated that levels of these compounds in human milk are quite low (*52*). Thus, direct transfer during pregnancy may result in a reduction in the quantities transferred for subsequent pregnancies. Future studies are needed to examine the extent of maternal−fetal transfer of these compounds.

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Literature Cited

(1) Kissa, E. *Fluorinated Surfactants and Repellents*, 2nd ed.; Marcel Dekker, Inc.: New York, 2001.

(2) 3M Company. *Fluorochemical use, distribution, and release overview*; EPA Docket # OPPT-2002-0043; 1999.

(3) Kannan, K.; Corsolini, S.; Falandysz, J.; Fillmann, G.; Kumar, K. S.; Loganathan, B. G.; Mohd, M. A.; Olivero, J.; Van, W. N.; Yang, J. H.; Aldoust, K. M. Perfluorooctanesulfonate and related fluorochemicals in human blood from several countries. *Environ. Sci. Technol.* **2004**, *38* (17), 4489−4495.

(4) Giesy, J. P.; Kannan, K. Global distribution of perfluorooctane sulfonate in wildlife. *Environ. Sci. Technol.* **2001**, *35* (7), 1339−1342.

(5) Calafat, A. M.; Kuklenyik, Z.; Caudill, S. P.; Reidy, J. A.; Needham, L. L. Perfluorochemicals in pooled serum samples from United States residents in 2001 and 2002. *Environ. Sci. Technol.* **2006**, *40* (7), 2128−2134.

(6) Calafat A. M.; Kuklenyik Z.; Reidy, J. A.; Caudill, S. P.; Tully, J. S.; Needham L. L. Serum Concentrations of 11 Polyfluoroalkyl Compounds in the U.S. Population: Data from the National Health and Nutrition Examination Survey (NHANES) 1999−2000. *Environ. Sci. Technol.* **2007**, *41* (7), 2237−2242.

(7) 3M Company. *Letter to EPA re: phase-out plan for POSF-based products*; EPA Docket 2002-0043-0009; St. Paul, MN, 2000.

(8) DuPont Commits to Aggressive PFOA Emissions Reduction Program Announced by the U.S. Environmental Protection Agency (press release); http://www2.dupont.com/Media_Center/en_US/news_releases/2006/article20060125d.html (last update January 25, 2006).

(9) Shipley, J. M.; Hurst, C. H.; Tanaka, S. S.; DeRoos, F. L.; Butenhoff, J. L.; Seacat, A. M.; Waxman, D. J. Trans-activation of PPARalpha and induction of PPARalpha target genes by perfluorooctane-based chemicals. *Toxicol. Sci.* **2004**, *80* (1), 151−160.

(10) Seacat, A. M.; Thomford, P. J.; Hansen, K. J.; Olsen, G. W.; Case, M. T.; Butenhoff, J. L. Subchronic toxicity studies on perfluorooctanesulfonate potassium salt in cynomolgus monkeys. *Toxicol. Sci.* **2002**, *68* (1), 249−264.

(11) Haughom, B.; Spydevold, O. The mechanism underlying the hypolipemic effect of perfluorooctanoic acid (PFOA), perfluorooctane sulphonic acid (PFOSA), and clofibric acid. *Biochim. Biophys. Acta* **1992**, *1128* (1), 65−72.

(12) Thibodeaux, J. R.; Hanson, R. G.; Rogers, J. M.; Grey, B. E.; Barbee, B. D.; Richards, J. H.; Butenhoff, J. L.; Stevenson, L. A.; Lau, C. Exposure to perfluorooctane sulfonate during pregnancy in rat and mouse. I: maternal and prenatal evaluations. *Toxicol. Sci.* **2003**, *74* (2), 369−381.

(13) Austin, M. E.; Kasturi, B. S.; Barber, M.; Kannan, K.; MohanKumar, P. S.; MohanKumar, S. M. Neuroendocrine effects of perfluorooctane sulfonate in rats. *Environ. Health Perspect.* **2003**, *111* (12), 1485−1489.

(14) Lau, C.; Thibodeaux, J. R.; Hanson, R. G.; Rogers, J. M.; Grey, B. E.; Stanton, M. E.; Butenhoff, J. L.; Stevenson, L. A. Exposure to perfluorooctane sulfonate during pregnancy in rat and mouse. II: postnatal evaluation. *Toxicol. Sci.* **2003**, *74* (2), 382−392.

(15) Luebker, D. J.; York, R. G.; Hansen, K. J.; Moore, J. A.; Butenhoff, J. L. Neonatal mortality from in utero exposure to perfluorooctanesulfonate (PFOS) in Sprague-Dawley rats: dose-response, and biochemical and pharamacokinetic parameters. *Toxicology* **2005**, *215* (1−2), 149−169.

(16) Luebker, D. J.; Case, M. T.; York, R. G.; Moore, J. A.; Hansen, K. J.; Butenhoff, J. L. Two-generation reproduction and cross-foster studies of perfluorooctanesulfonate (PFOS) in rats. *Toxicology* **2005**, *215* (1−2), 126−148.

(17) OECD. *Hazard Assessment of Perfluorooctane Sulfonate (PFOS) and its Salts*; ENV/JM/RD(2002)17/FINAL; Organization for Economic Cooperation and Development; Environment Directorate; Joint Meeting of the Chemicals Committee and the Working Party on Chemicals, Pesticides, and Biotechnology: Paris, 2002.

(18) Kennedy, G. L., Jr.; Butenhoff, J. L.; Olsen, G. W.; O'Connor, J. C.; Seacat, A. M.; Perkins, R. G.; Biegel, L. B.; Murphy, S. R.; Farrar, D. G. The toxicology of perfluorooctanoate. *Crit. Rev. Toxicol.* **2004**, *34* (4), 351−384.

(19) Ikeda, T.; Aiba, K.; Fukuda, K.; Tanaka, M. The induction of peroxisome proliferation in rat liver by perfluorinated fatty acids, metabolically inert derivatives of fatty acids. *J. Biochem. (Tokyo)* **1985**, *98* (2), 475−482.

(20) Kudo, N.; Kawashima, Y. Toxicity and toxicokinetics of perfluorooctanoic acid in humans and animals. *J. Toxicol. Sci.* **2003**, *28* (2), 49−57.

(21) U.S. Environmental Protection Agency Science Advisory Board (SAB). *SAB Review of EPA's Draft Risk Assessment of Potential Human Health Effects Associated with PFOA and Its Salts*; EPA-SAB-06-006; Washington, DC, 2006.

(22) Butenhoff, J. L.; Kennedy, G. L., Jr.; Frame, S. R.; O'Connor, J. C.; York, R. G. The reproductive toxicology of ammonium perfluorooctanoate (APFO) in the rat. *Toxicology* **2004**, *196* (1−2), 95−116.

(23) Lau, C.; Butenhoff, J. L.; Rogers, J. M. The developmental toxicity of perfluoroalkyl acids and their derivatives. *Toxicol. Appl. Pharmacol.* **2004**, *198* (2), 231−241.

(24) Lau, C.; Thibodeaux, J. R.; Hanson, R. G.; Narotsky, M. G.; Rogers, J. M.; Lindstrom, A. B.; Strynar, M. J. Effects of perfluorooctanoic acid exposure during pregnancy in the mouse. *Toxicol. Sci.* **2006**, *90* (2), 510−518.

(25) Olsen, G.; Ehresman, D.; Froehlich, J.; Burris J.; Butenhoff, J. Evaluation of the half-life ($T_{1/2}$) of elimination of perfluorooctanesulfonate (PFOS), perfluorohexanesulfonate (PFHS) and perfluorooctanoate (PFOA) from human serum. Presented at *FLUOROS: An international symposium on fluorinated alkyl organics in the environment*; August 19, 2005.

(26) Jones, P. D.; Hu, W.; De Coen, W.; Newsted, J. L.; Giesy, J. P. Binding of perfluorinated fatty acids to serum proteins. *Environ. Toxicol. Chem.* **2003**, *22* (11), 2639−2649.

(27) Luebker, D. J.; Hansen, K. J.; Bass, N. M.; Butenhoff, J. L.; Seacat, A. M. Interactions of fluorochemicals with rat liver fatty acid-binding protein. *Toxicology* **2002**, *176* (3), 175−185.

(28) Vanden Heuvel, J. P.; Kuslikis, B. I.; Peterson, R. E. Covalent binding of perfluorinated fatty acids to proteins in the plasma, liver and testes of rats. *Chem. Biol. Interact.* **1992**, *82* (3), 317−328.

(29) Olsen, G. W.; Church, T. R.; Miller, J. P.; Burris, J. M.; Hansen, K. J.; Lundberg, J. K.; Armitage, J. B.; Herron, R. M.; Medhdizadehkashi, Z.; Nobiletti, J. B.; O'Neill, E. M.; Mandel, J. H.; Zobel, L. R. Perfluorooctanesulfonate and other fluorochemicals in the serum of American Red Cross adult blood donors. *Environ. Health Perspect.* **2003**, *111* (16), 1892−1901.

(30) Olsen, G. W.; Church, T. R.; Hansen, K. J.; Burris, J. M.; Butenhoff, J. L.; Mandel, J. H.; Zobel, L. R. Quantitative evaluation of perfluorooctanesulfonate (PFOS) and other fluorochemicals in the serum of children. *J. Children's Health* **2004**, *2* (1), 53−76.

(31) Olsen, G. W.; Church, T. R.; Larson, E. B.; van Belle, G.; Lundberg, J. K.; Hansen, K. J.; Burris, J. M.; Mandel, J. H.; Zobel, L. R. Serum concentrations of perfluorooctanesulfonate and other fluorochemicals in an elderly population from Seattle, Washington. *Chemosphere* **2004**, *54* (11), 1599−1611.

(32) Inoue, K.; Okada, F.; Ito, R.; Kato, S.; Sasaki, S.; Nakajima, S.; Uno, A.; Saijo, Y.; Sata, F.; Yoshimura, Y.; Kishi, R.; Nakazawa, H. Perfluorooctane sulfonate (PFOS) and related perfluorinated compounds in human maternal and cord blood samples: assessment of PFOS exposure in a susceptible population during pregnancy. *Environ. Health Perspect.* **2004**, *112* (11), 1204−1207.

(33) Tittlemier, S.; Ryan, J. J.; Van Oostdam, J. Presence of anionic organic compounds in serum collected from northern Canadian populations. *Organohalogen Compd.* **2004**, *66*, 4009−4014.

(34) Midasch, O.; Drexler, H.; Hart, N.; Beckmann, M. W.; Angerer, J. Transplacental exposure of neonates to perfluorooctanesulfonate and perfluorooctanoate: a pilot study. *Int. Arch. Occup. Environ. Health* [Online early access]. 10.1007/s00420-006-0165-9. Published Online: Jan 12, 2007.

(35) Witter, F. R.; Ten Broeck, J.; Fox, H. E. A new device for safer collection of postpartum cord blood. *Int. J. Gynaecol. Obstet.* **2001**, *72* (3), 259−260.

(36) U.S. Centers for Disease Control and Prevention (CDC). *Third National Report on Human Exposure to Environmental Chemicals*; NCEH Pub. No. 05-0570; Department of Health and Human Services, Centers for Disease Control and Prevention, National Center for Environmental Health, Division of Laboratory Sciences: Atlanta, GA, 2005.

(37) Kuklenyik, Z.; Needham, L. L.; Calafat, A. M. Measurement of 18 perfluorinated organic acids and amides in human serum using on-line solid-phase extraction. *Anal. Chem.* **2005**, *77* (18), 6085−6091.

(38) Bernert, J. T., Jr.; Turner, W. E.; Pirkle, J. L.; Sosnoff, C. S.; Akins, J. R.; Waldrep, M. K.; Ann, Q.; Covey, T. R.; Whitfield, W. E.;

Gunter, E. W.; Miller, B. B.; Patterson, D. G., Jr.; Needham, L. L.; Hannon, W. H.; Sampson, E. J. Development and validation of sensitive method for determination of serum cotinine in smokers and nonsmokers by liquid chromatography/atmospheric pressure ionization tandem mass spectrometry. *Clin. Chem.* **1997**, *43* (12), 2281−2291.

(39) Hornung, R. W.; Reed, L. D. Estimation of average concentration in the presence of nondetectable values. *Appl. Occup. Environ. Hyg.* **1990**, *5* (1), 46−51.

(40) Dinglasan-Panlilio, M. J.; Mabury, S. A. Significant residual fluorinated alcohols present in various fluorinated materials. *Environ. Sci. Technol.* **2006**, *40* (5), 1447−1453.

(41) Begley, T. H.; White, K.; Honigfort, P.; Twaroski, M. L.; Neches, R.; Walker, R. A. Perfluorochemicals: potential sources of and migration from food packaging. *Food Addit. Contam.* **2005**, *22* (10), 1023−1031.

(42) Kubwabo, C.; Stewart, B.; Zhu, J.; Marro, L. Occurrence of perfluorosulfonates and other perfluorochemicals in dust from selected homes in the city of Ottawa, Canada. *J. Environ. Monit.* **2005**, *7* (11), 1074−1078.

(43) Shoeib, M.; Harner, T.; Wilford, B. H.; Jones, K. C.; Zhu, J. Perfluorinated sulfonamides in indoor and outdoor air and indoor dust: occurrence, partitioning, and human exposure. *Environ. Sci. Technol.* **2005**, *39* (17), 6599−6606.

(44) Saito, N.; Harada, K.; Inoue, K.; Sasaki, K.; Yoshinaga, T.; Koizumi, A. Perfluorooctanoate and perfluorooctane sulfonate concentrations in surface water in Japan. *J. Occup. Health* **2004**, *46* (1), 49−59.

(45) Sinclair, E.; Mayack, D. T.; Roblee, K.; Yamashita, N.; Kannan, K. Occurrence of perfluoroalkyl surfactants in water, fish, and birds from New York State. *Arch. Environ. Contam. Toxicol.* **2006**, *50* (3), 398−410.

(46) Kannan, K.; Tao, L.; Sinclair, E.; Pastva, S. D.; Jude, D. J.; Giesy, J. P. Perfluorinated compounds in aquatic organisms at various trophic levels in a Great Lakes food chain. *Arch. Environ. Contam Toxicol.* **2005**, *48* (4), 559−566.

(47) Smithwick, M.; Mabury, S. A.; Solomon, K. R.; Sonne, C.; Martin, J. W.; Born, E. W.; Dietz, R.; Derocher, A. E.; Letcher, R. J.; Evans, T. J.; Gabrielsen, G. W.; Nagy, J.; Stirling, I.; Taylor, M. K.; Muir, D. C. Circumpolar study of perfluoroalkyl contaminants in polar bears (*Ursus maritimus*). *Environ. Sci. Technol.* **2005**, *39* (15), 5517−5523.

(48) Taniyasu, S.; Kannan, K.; Horii, Y.; Hanari, N.; Yamashita, N. A survey of perfluorooctane sulfonate and related perfluorinated organic compounds in water, fish, birds, and humans from Japan. *Environ. Sci. Technol.* **2003**, *37* (12), 2634−2639.

(49) Falandysz, J.; Taniyasu, S.; Gulkowska, A.; Yamashita, N.; Schulte-Oehlmann, U. Is fish a major source of fluorinated surfactants and repellents in humans living on the Baltic Coast? *Environ. Sci. Technol.* **2006**, *40* (3), 748−751.

(50) Brenner, H.; Loomis, D. Varied forms of bias due to nondifferential error in measuring exposure. *Epidemiology* **1994**, *5* (5), 510−517.

(51) Butenhoff, J. L.; Gaylor, D. W.; Moore, J. A.; Olsen, G. W.; Rodricks, J.; Mandel, J. H.; Zobel, L. R. Characterization of risk for general population exposure to perfluorooctanoate. *Regul. Toxicol. Pharmacol.* **2004**, *39* (3), 363−380.

(52) Kärrman, A.; Ericson, I.; van Bavel, B.; Darnerud, P. O.; Aune, M.; Glynn, A.; Lignell, S.; Lindström, G. Exposure of Perfluorinated Chemicals through Lactation: Levels of Matched Human Milk and Serum and a Temporal Trend, 1996−2004, in Sweden. *Environ. Health Perspect.* **2007**, *115* (2), 226−230.

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