The Association Between Perfluoroalkyl Substances and Lipids in Cord Blood

Miranda J. Spratlen,[1] Frederica P. Perera,[1] Sally Ann Lederman,[2] Morgan Robinson,[3] Kurunthachalam Kannan,[3,4] Julie Herbstman,[1,*] and Leonardo Trasande[5,6,7,*]

[1]Columbia Center for Children's Environmental Health, Department of Environmental Health Sciences, Columbia University Mailman School of Public Health, New York, New York, 10032; [2]Department of Population and Family Health, Columbia University Mailman School of Public Health, New York, New York, 10032; [3]Wadsworth Center, New York State Department of Health, Albany, New York, 12201; [4]Department of Environmental Health Sciences, School of Public Health, State University of New York at Albany, Albany, New York, 12144; [5]Department of Pediatrics, New York University School of Medicine, New York, New York, 10016; [6]Department of Environmental Medicine, New York University School of Medicine, New York, New York, 10016; and [7]Department of Population Health, New York University School of Medicine, New York, New York, 10016

ORCiD numbers: 0000-0002-4486-4658 (M. J. Spratlen).

Introduction: Perfluoroalkyl substances (PFAS) were among various persistent organic pollutants suspected to have been released during the collapse of the World Trade Center (WTC) on 9/11/2001. Evidence suggests that PFAS may have cardiometabolic effects, including alterations in lipid profiles. This study evaluated the association between cord blood PFAS and lipids in a population prenatally exposed to the WTC disaster.

Study Population: 222 pregnant women in the Columbia University WTC birth cohort enrolled between December 13, 2001 and June 26, 2002 at hospitals located near the WTC site: Beth Israel, St. Vincent's, and New York University Downtown.

Methods: We evaluated the association between 5 cord blood PFAS—perfluorooctane sulfonate (PFOS), perfluorooctanoic acid (PFOA), perfluorohexanesulfonic acid (PFHxS), perfluorononanoic acid (PFNA), perfluorodecane sulfonate (PFDS)—and cord blood lipids (total lipids, total cholesterol, triglycerides).

Results: Median (interquartile range [IQR]) concentrations of PFAS were 6.32 (4.58–8.57), 2.46 (1.77–3.24), 0.38 (0.25–0.74), 0.66 (0.48–0.95) and 0.11 (0.09–0.16) ng/mL for PFOS, PFOA, PFNA, PFHxS, and PFDS, respectively. Median (IQR) for lipids were 59.0 (51.5–68.5) mg/dL for total cholesterol, 196.5 (170.5–221.2) mg/dL for total lipids and 33.1 (24.2–43.9) mg/dL for triglycerides. In fully adjusted models, several PFAS were associated with higher lipid levels, including evidence of a strong linear trend between triglycerides and both PFOA and PFHxS.

Conclusions: Findings support previous evidence of an association between PFAS exposure and altered lipid profiles and add novel information on this relationship in cord blood, as well as for an understudied PFAS, PFDS (***J Clin Endocrinol Metab* XX: 0-0, 2019**).

Key Words: perfluoroalkyl substances, cord blood, World Trade Center disaster, lipids

ISSN Print 0021-972X ISSN Online 1945-7197
Printed in USA

Received 12 March 2019. Accepted 17 September 2019.
First Published Online 19 September 2019.

*Co-last authors.

Abbreviations: EPA, U.S. Environmental Protection Agency; PFAS, perfluoroalkyl substances; PFBS, perfluorobutanesulfonic acid; PFDA, perfluorodecanoic acid; PFDoDA, perfluorododecanoate; PFHpA, perfluoroheptanoic acid; PFDS, perfluorodecane sulfonate; PFHxA, perfluorohexanoic acid; PFHxS, perfluorohexanesulfonic acid; PFNA, perfluorononanoic acid; PFOA, perfluorooctanoic acid; PFOS, perfluorooctane sulfonate; PFOSA, perfluorooctane sulfonamide; PFUnDA, perfluoroundecanoic acid; SPE, solid phase extraction; WTC, World Trade Center.

Perfluoroalkyl substances (PFAS) are persistent and ubiquitous environmental contaminants with surfactant and oil-repelling properties that have led to their widespread use in commercial and industrial products, including firefighting foam, carpets, food packaging, clothing and nonstick cookware (1). Exposure in humans occurs mostly through contaminated food and water; however, dust and fumes from treated products in homes and offices have also been identified as additional routes of exposure (2, 3). National trends suggest reductions in exposure among adults over the past 2 decades (4, 5) for the 2 most widely produced PFAS, perfluorooctanoic acid (PFOA) and perfluorooctane sulfonate (PFOS), with more recent reductions also observed for less common PFAS, including perfluorohexanesulfonic acid (PFHxS), perfluorononanoic acid (PFNA), and perfluorodecane sulfonic acid (PFDS) (5).

Still, PFAS remain a global public health priority; in 2016, the U.S. Environmental Protection Agency (EPA) issued a Lifetime Health Advisory (6) for PFAS in drinking water; and in 2010 and 2016, measures were adopted by the European Union to monitor PFAS in food (7) and regulate PFAS in drinking water (8), respectively. Their persistence, wide distribution, and bioaccumulation in the environment, as well as their potential to cause human health effects, has led to their designation by the EPA as an "emerging contaminant" (1). Indeed, epidemiological evidence suggests even low-level exposures to PFAS may be associated with adverse health effects, including increases in blood lipids and liver enzymes, as well as cancer, immune suppression, and thyroid disorders (9, 10). Studies have also shown that PFAS can cross the placental barrier and may cause reductions in fetal growth (11, 12).

We recently found that pregnant women who lived or worked within 2 miles of the World Trade Center (WTC) during the 4-week period following the attack on 9/11/2001, had higher cord blood PFAS levels than pregnant women who were located farther from the disaster (13). Potential PFAS use in office buildings, including carpeting and furniture, as well as its being an active ingredient in aqueous firefighting foam, provide possible explanations for proximity to the WTC disaster having an association with PFAS exposure (1). These results identified a potentially vulnerable population and emphasized the need for more research to evaluate possible health effects resulting from in utero exposure to the disaster. Cardiovascular disease is the leading cause of death both nationally (14) and globally (15) and has been shown to have some prenatal origins (16). Mounting epidemiological evidence supports a relationship between PFAS exposure and cardiometabolic outcomes, including increases in triglycerides (17–19) and total cholesterol (4, 17, 18, 20–25), in both adults and children. Because of the sensitivity of the prenatal period and the potential for exposures during this time to influence postnatal health, recent studies have also evaluated the relationship between maternal PFAS exposure and maternal lipids (26, 27), as well as lipid profiles in their offspring years later (28–30), with inconsistent findings. To our knowledge, this is the first study to evaluate the relationship between cord blood PFAS levels and cord blood lipids, including triglycerides, total cholesterol, and total lipids. Because PFAS appear to have variable transfer efficiencies from maternal blood to cord blood (31–33), measuring both the PFAS and lipid variables in cord blood may reflect the exposure-outcome relationship more accurately. Furthermore, our analyses were conducted in the Columbia University WTC birth cohort, a vulnerable population potentially at higher risk for adverse health outcomes resulting from proximity to the WTC disaster during gestation.

Methods

Study population

The study population was drawn from a Columbia University birth cohort designed to evaluate the effects of WTC exposures on pregnancy outcomes and child development. Detailed methods have been described previously (34). Briefly, 329 women with singleton pregnancies were enrolled between December 13, 2001 and June 26, 2002 at 1 of 3 hospitals located near the WTC site: Beth Israel, St. Vincent's, and New York University Downtown. Eligibility requirements included: maternal age from 18 to 39 years, had not smoked (> 1 cigarette at any time) during pregnancy, self-report of no diabetes, hypertension, HIV infection or AIDS, and no use of illegal drugs in the last year. Participants provided at least 1 blood sample (maternal blood at the time of delivery and/or cord blood), access to their medical record and their newborn's medical record, and completion of a 30- to 45-minute interview after delivery. Participants who were missing data on cord blood PFAS (n = 82) or lipids (n = 9), race (n = 12), body mass index (BMI) (n = 3), and parity (n = 1) were excluded, resulting in a sample size of 222 participants for analyses (Fig. 1).

Sociodemographic variables

The postpartum interview was administered at the hospital, after delivery and prior to discharge, in the woman's preferred or native language (English, Spanish, or Chinese). Information on maternal education, date of birth, race, parity, marital status, and family smoking exposure was collected through the interview. Maternal pre-pregnancy BMI was calculated using weight in kilograms divided by height in meters squared (kg/m^2), both abstracted from the participants' medical chart. In the case of missing height (n = 36) or weight (n = 49) data from the medical record, self-reported information on these

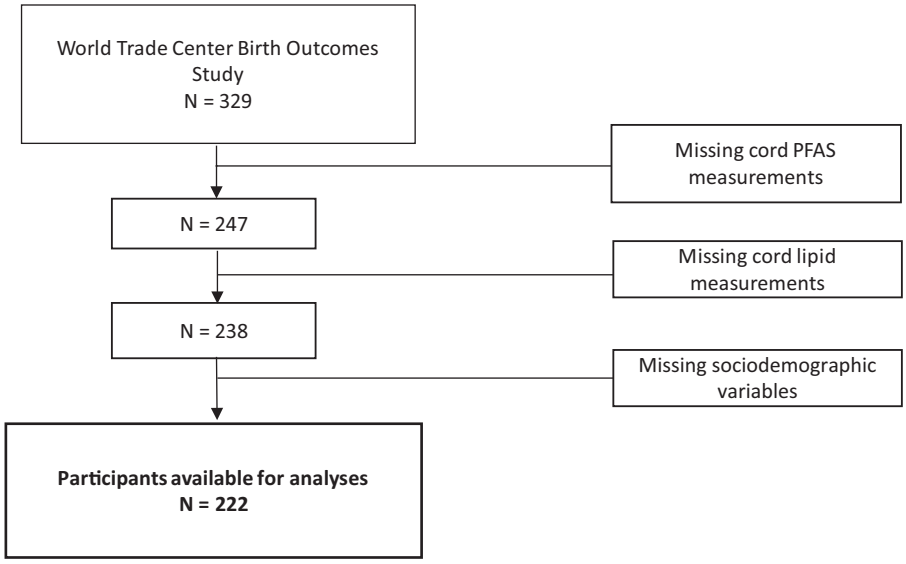

Figure 1. Study flow diagram. Abbreviations: PFAS, perfluoroalkyl substances.

variables from the hospital interview were used. Among participants with both self-reported and medical record weight and height, correlations were very high (r = 0.99 and r = 0.91, respectively). Child sex and date of birth were abstracted from the child's medical record. Gestational age in days was also abstracted from the medical record (if missing (n = 15), the date of last menstrual period from interview minus child's date of birth was used). Maternal age at delivery was determined by subtracting the child's date of birth from the mother's date of birth. Institutional review board approval was obtained before enrollment began and all women gave written informed consent before delivery. The Centers for Disease Control and Prevention (CDC), which measured lipids, was determined not to be engaged in human-subjects research because no personally identifiable information was made available to CDC researchers.

Cord blood collection and measurements

Blood samples from the umbilical cord were collected at the time of delivery, transported to Columbia University laboratory facilities in Northern Manhattan and processed within hours of collection. The buffy coat, packed red blood cells, and plasma were separated and stored at −70°C.

PFAS measurements. Twelve PFAS [PFOS, PFOA, PFHxS, PFDS, PFNA, perfluorobutanesulfonic acid (PFBS), perfluorooctane sulfonamide (PFOSA), perfluorohexanoic acid (PFHxA), perfluoroheptanoic acid (PFHpA), perfluorodecanoic acid (PFDA), perfluoroundecanoic acid (PFUnDA) and perfluorododecanoate (PFDoDA)] were measured in cord blood using a solid phase extraction procedure and high-performance liquid chromatograph interfaced with an electrospray tandem mass spectrometer, at the New York State Department of Health Wadsworth Center Laboratory, using methods similar to prior studies (35, 36). Internal standards for 13C-labeled PFAS were added into plasma samples prior to the addition of reagents for extraction (37). Solvents and method blanks (blinded to the laboratory) were tested for the presence of the PFAS. Target chemicals were not found in procedural blanks at concentrations above the limits of quantification (LOQs). The LOQs of target chemicals ranged from 0.08 to 0.20 ng/mL. A standard reference material from the National Institute of Standards and Technology (NIST) was analyzed with every batch of 50 samples and recoveries of target chemicals were between 90% and 115% of the certified values. Recoveries of target chemicals passed through the entire analytical procedure ranged between 100% and 124%. Quantification was by isotope dilution and target chemicals were monitored by multiple reaction monitoring mode under negative ionization.

Lipid measurements. Plasma triglycerides and total cholesterol were measured on a Hitachi 704 Analyzer at the CDC's Persistent Organic Pollutants Biomonitoring Laboratory at the National Center for Environmental Health using commercially available test kits from Roche Diagnostics Corp. Cholesterol was measured enzymatically using the Cholesterol High Performance reagent (cat. no. 704036), Roche Diagnostics). Triglycerides were analyzed enzymatically simultaneously with cholesterol, using reagents from the same manufacturer (Triglycerides/GPO, cat. no. 1488872). Triglyceride blanks were measured in CDC surveillance materials using the same reagent, but without lipase. Total lipids were determined from total triglycerides and total cholesterol, as described in statistical methods.

Statistical analyses

All statistical analyses were conducted in R software (version 3.5.1; R Project for Statistical Computing). PFAS assessment was restricted to compounds quantified in ≥ 50% of samples (PFOS, PFOA, PFNA, PFHxS and PFDS). Both PFOA and PFOS were detected in 100% of samples. One sample (< 1%) was below the LOQ (0.08 ng/mL) for PFHxS, 29 samples (13%) were below the LOQ (0.20 ng/mL) for PFNA, and 6 (3%) were below the LOQ (0.08) for PFDS. In accord with published practices (38), samples < LOQ were imputed as the LOQ divided by √2.

Total serum lipids are normally calculated using an equation known as the "Phillips" or "short" equation (39), which estimates the contribution of phospholipids to total lipids using triglycerides and total cholesterol. This formula, however, was derived based on adult serum collected in the 1980s. By actually measuring phospholipid levels in 100 cord blood samples, our research group determined that this formula underestimated total lipids when used on cord samples. Using these data, our group reconstructed the short formula to more accurately reflect total lipids in cord blood (manuscript under review). Therefore, for our total lipid analyses, we used this modified formula: Total Cord Lipids = (2.657 x Total Cord Cholesterol) + 0.268 + Cord Triglycerides. In a sensitivity analysis, we used the standard formula for total serum lipids: Total Lipids = (2.27 × Total Cholesterol) + 0.623 + Triglycerides. Total serum lipids, total cholesterol and triglycerides were log-transformed in all linear regression analyses to account for right-skewed distributions.

Medians (IQR) and percentages were used to describe sociodemographic variables and lipids (total lipids, total cholesterol, and triglycerides) in the study population overall and by high and low PFAS exposure. We identified low and high PFAS exposure through principal component analyses (cutting our cumulative PFAS exposure variable at equal to or below the median and above the median, to reflect low and high exposure, respectively). We used principal component analyses as a data reduction technique to extract a smaller number of latent variables summarizing most of the variance of the observed PFAS measures, as has been done in previous PFAS analyses (40). We included all principal factors with eigenvalues ≥1.0 (41). Differences between participants across exposure categories were calculated using Mann-Whitney U and chi-square tests for continuous and categorical variables, respectively. Spearman correlations between log-transformed PFAS variables and log-transformed lipids were explored using a correlation matrix. Linear regression models were used to evaluate the association between log-transformed PFAS variables and log-transformed lipid variables and report percent differences in lipids by percent increase in PFAS concentration. To better understand linearity of associations, models were also run evaluating the association between quartiles of PFAS variables and log-transformed lipid variables; coefficients were exponentiated and reported as geometric mean ratios. To formally assess linear trends, *P*-trend values were obtained by regressing lipids on PFAS quartile variables modeled continuously. Relationships were visually evaluated using restricted cubic splines.

Models were adjusted for variables selected *a priori* based on previous literature, including: child sex; maternal race (black, white, Asian, Native American/other); maternal pre-pregnancy BMI (underweight: BMI < 18.5 kg/m^2; normal: BMI ≥ 18.5 and < 25 kg/m^2; overweight/obese: BMI ≥ 25 kg/m^2); maternal education (< high school degree, high school degree, > high school degree); home smoking exposure (no reported family member smoking in household, any reported family member smoking in household); marital status (not married, married); and parity (primiparous, multiparous). Due to the reported association between gestational age and cord lipids, as this analysis was conducted in cord blood, we also adjusted models for gestational age to improve our prediction of lipid concentrations.

Results

Participant characteristics

Principal component analyses showed that the majority of PFAS exposure variance (54%) was captured in the first principal component and reflected higher levels of all 5 PFAS (Table 1). The second principal component captured 27% of PFAS exposure variance and reflected higher PFOA and PFHxS levels. Principal component 1 was split at below or equal to the median to reflect low PFAS exposure and above the median to reflect high PFAS exposure. Differences in study participant characteristics across high and low PFAS exposure categories were only significant by race and education (Table 2). Participants with high PFAS exposure were more likely to have less than a high school degree (P = 0.02) and to be of Asian ethnicity (P = 0.003). Participants with high PFAS exposure were less likely to have greater than a high school degree and to be of white ethnicity.

The median age of participants in this study at enrollment was 30.6 years (Table 2). Roughly the same percentage of participants were Asian (40.5%) and white (39.6%), with smaller proportions reporting being of black (15.3%) or other (4.5%) race. Most participants were married (83.8%) and reported no smoking in the household (82%). The majority of participants had greater than a high school degree (62.2%), with 17.1% reporting only a high school degree and 20.7% reporting less than a high school degree. Most mothers had a normal pre-pregnancy BMI (71.6%), 18.5% were overweight or obese, and just 9.9% were underweight. Slightly more participants were primiparous (55.0%) and pregnant with a female child (54.5%). Median (IQR) cord lipid levels were 59.0 (51.5–68.5) mg/dL for total cholesterol, 196.5 (170.5–221.2) mg/dL for

Table 1. Principal Components of Perfluoroalkyl Substances (PFAS) in Cord Blood

PFAS	Principal Component 1	Principal Component 2
Standard deviation	1.63	1.16
Proportion of variance	53.7%	27.0%
Weight for PFOS	0.57	0.02
Weight for PFOA	0.31	0.57
Weight for PFNA	0.51	-0.33
Weight for PFHxS	0.29	0.63
Weight for PFDS	0.48	-0.41

Abbreviations: PFDS, perfluorodecane sulfonate; PFHxS, perfluorohexanesulfonic acid; PFNA, perfluorononanoic acid; PFOA, perfluorooctanoic acid; PFOS, perfluorooctane sulfonate.

Table 2. Participant Characteristics of Study Population Overall and by PFAS Exposure

Variable	Median (IQR) or N (%)			*P* value
	Overall	Low PFAS*	High PFAS*	
N (%)	222	111 (50)	111 (50)	
Maternal age (years)	30.6 (27.1–34.5)	29.5 (26.4–33.9)	31.5 (27.6–34.6)	0.16
Race, n (%)				
Black	34 (15.3)	23 (20.7)	11 (9.9)	0.003
White	88 (39.6)	51 (45.9)	37 (33.3)	
Asian	90 (40.5)	32 (28.8)	58 (52.3)	
Other	10 (4.5)	5 (4.5)	5 (4.5)	
Pre-pregnancy BMI, n (%)				
Underweight	22 (9.9)	10 (9)	12 (10.8)	0.29
Normal	159 (71.6)	76 (68.5)	83 (74.8)	
Overweight/Obese	41 (18.5)	25 (22.5)	16 (14.4)	
Parity, n (%)				
Primiparous	122 (55.0)	62 (55.9)	60 (54.1)	0.89
Multiparous	100 (45.0)	49 (44.1)	51 (45.9)	
Marital status, n (%)				
Single	36 (16.2)	18 (16.2)	18 (16.2)	1.00
Married	186 (83.8)	93 (83.8)	93 (83.8)	
Education, n (%)				
< High school degree	46 (20.7)	15 (13.5)	31 (27.9)	0.02
High school degree	38 (17.1)	19 (17.1)	19 (17.1)	
> High school degree	138 (62.2)	77 (69.4)	61 (55.0)	
Family smoking status, n (%)				
No family smoking	182 (82.0)	88 (79.3)	94 (84.7)	0.38
Any family smoking	40 (18.0)	23 (20.7)	17 (15.3)	
Child sex, n (%)				
Female	121 (54.5)	64 (57.7)	57 (51.4)	0.42
Male	101 (45.5)	47 (42.3)	54 (48.6)	
Cord cholesterol (mg/dL), median (IQR)	59.0 (51.5–68.5)	58.9 (51.6–67.7)	59.1 (51.5–69.2)	0.64
Cord total lipids (mg/dL), median (IQR)	196.5 (170.5–221.2)	192.9 (170.7–218.7)	199.4 (170.2–222.2)	0.43
Cord triglycerides (mg/dL), median (IQR)	33.1 (24.2–43.9)	32.9 (24.9–42.9)	34.6 (23.7–45.4)	0.75

*Low and High PFAS defined as ≤ and > median of PFAS principal component 1 (summary PFAS measure reflective of higher exposure to all PFAS), respectively. Abbreviations: BMI, body mass index; IQR, interquartile range; PFAS, perfluoroalkyl substances.

total lipids, and 33.1 (24.2–43.9) mg/dL for triglycerides. Median (IQR) concentrations of cord PFAS were 6.32 (4.58–8.57) ng/mL for PFOS, 2.46 (1.77–3.24) ng/mL for PFOA, 0.38 (0.25–0.74) ng/mL for PFNA, 0.66 (0.48–0.95) ng/mL for PFHxS, and 0.11 (0.09–0.16) ng/mL for PFDS (Table 3).

Association between PFAS and lipids

All lipid variable concentrations were significantly ($P < 0.05$) and positively correlated with each other, ranging from moderate correlations between triglycerides and total cholesterol ($r = 0.19$) to high correlations between total cholesterol and total lipids ($r = 0.91$) (Fig. 2). All PFAS variables were also positively correlated with each other but not all significantly. PFOS and PFNA had the strongest correlation ($r = 0.70$) and were also significantly correlated with all other PFAS. PFOA and PFHxS were significantly correlated with all PFAS except PFDS. Correlations between PFAS and lipids ranged from very weak and not significant between PFDS and total lipids ($r = 0.01$) to moderate and significant between PFOA and triglycerides ($r = 0.30$).

In fully adjusted models, 1% higher PFOS was significantly associated with 0.067% (95% CI, 0.005–0.129) higher total lipids (Table 4). In quartile analyses, the p-trend was also significant (p-trend = 0.03) between PFOS and total lipids (Table 5). Higher PFOA was associated with higher total lipids [0.087% (95% CI, 0.021–0.153)] and higher triglycerides [0.256% (95% CI, 0.129–0.383)]. These associations were consistent in quartile analyses (p-trend = 0.04 for total lipids and p-trend = 0.001 for

Table 3. Median Concentrations (ng/mL) and Percent Above Quantification of Perfluoroalkyl Substances (PFAS) in Cord Blood

PFAS	LOQ	% Above LOQ	Median (IQR)
PFOS	0.20	100%	6.32 (4.58–8.57)
PFOA	0.08	100%	2.46 (1.77–3.24)
PFNA	0.20	87%	0.38 (0.25–0.74)
PFHxS	0.08	>99%	0.66 (0.48–0.95)
PFDS	0.08	97%	0.11 (0.09–0.16)

Abbreviations: LOQ, limit of quantification; IQR, interquartile range; PFDS, perfluorodecane sulfonate; PFHxS, perfluorohexanesulfonic acid; PFNA, perfluorononanoic acid; PFOA, perfluorooctanoic acid; PFOS, perfluorooctane sulfonate.

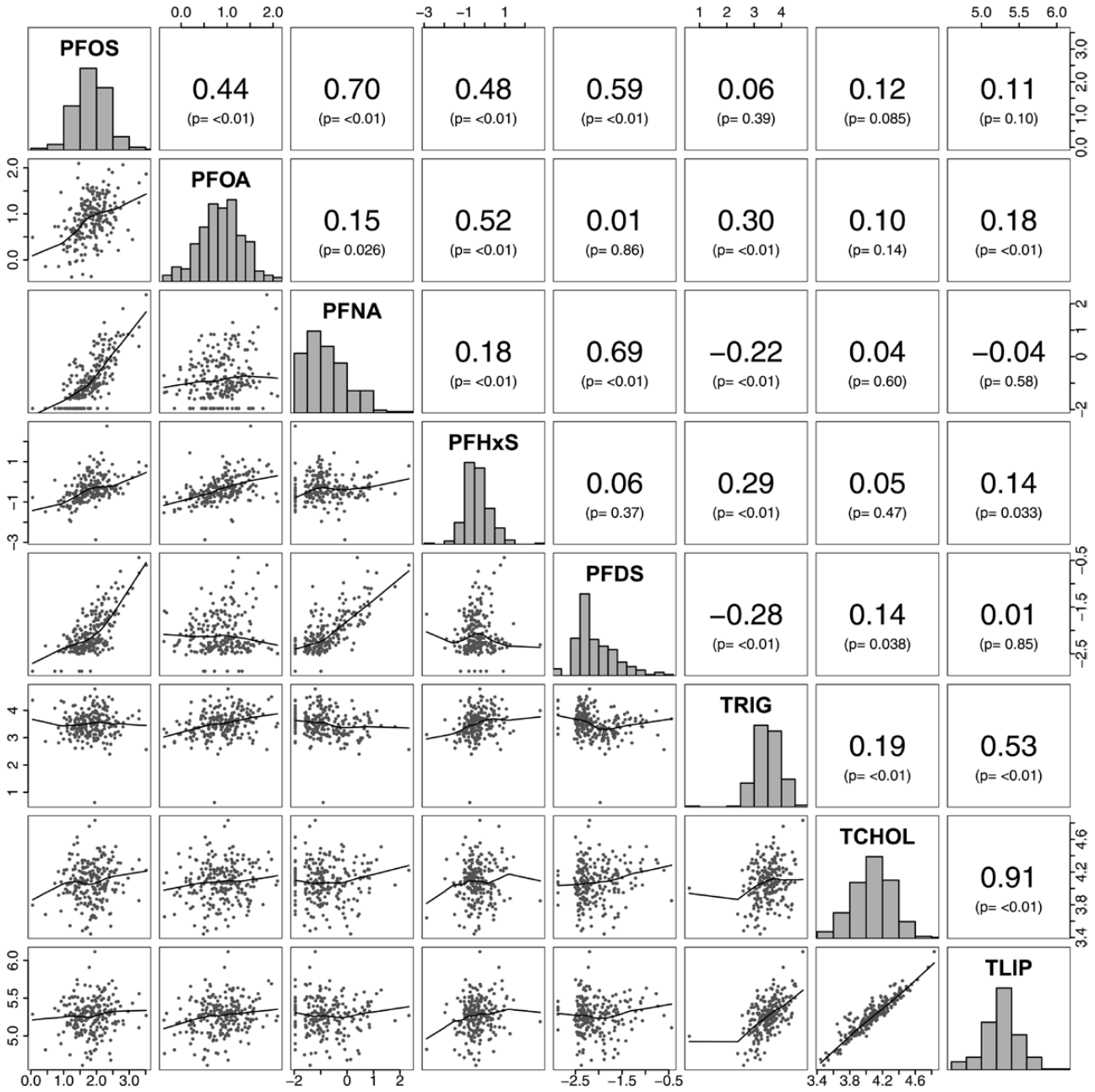

Figure 2. Correlation matrix of perfluroalkyl substances. Histograms of, and Spearman correlations between, log-transformed perfluroalkyl substances and lipids. Abbreviations: PFDS, perfluorodecane sulfonate; PFHxS, perfluorohexanesulfonic acid; PFNA, perfluorononanoic acid; PFOA, perfluorooctanoic acid; PFOS, perfluorooctane sulfonate; TCHOL, total cholesterol; TLIP, total lipids; TRIG, triglycerides.

triglycerides). Greater PFHxS concentrations were also associated with higher lipids and [0.051% (95% CI, 0.003–0.099)] and higher triglycerides [0.133% (95% CI, 0.038–0.226)]. In quartile analyses, the association between PFHxS and triglycerides was consistent (p-trend = 0.002). Higher PFDS was associated with higher total cholesterol [0.091% (95% CI, 0.006–0.177)]. This association was not consistent in quartile analyses (p-trend = 0.10), however, there was a significant negative trend between PFDS and triglycerides (p-trend = 0.04). PFNA was not associated with any lipid outcome. Consistent with quartile and linear regression models, visually, the strongest linear associations reflected in restricted cubic spline figures were between PFOA and PFHxS and triglycerides (Fig. 3). Results from a sensitivity analysis using the standard total lipids formula were consistent.

Principal component 1 (reflective of greater exposure to all 5 PFAS) was associated with higher total lipids [0.022 (95% CI, 0.002–0.042)] (Table 4). Principal component 2 (reflective of higher PFOA and PFHxS exposure) was also significantly associated with higher lipids [0.030 (95% CI, 0.00–0.061)]. In addition, consistent with both quartile and linear analyses, which showed associations between PFOA and PFHxS with higher triglycerides,

Table 4. Percent Difference in Lipids per 1% Higher PFAS and Principal Components (N = 222)

	Total Lipids	Total Cholesterol	Triglycerides
PFOS	0.067 (0.005, 0.129)	0.062 (-0.004, 0.127)	0.086 (-0.036, 0.209)
PFOA	0.087 (0.021, 0.153)	0.038 (-0.032, 0.109)	0.256 (0.129, 0.383)
PFNA	0.018 (-0.029, 0.066)	0.023 (-0.027, 0.074)	-0.03 (-0.124, 0.064)
PFHxS	0.051 (0.003, 0.099)	0.027 (-0.024, 0.078)	0.133 (0.038, 0.226)
PFDS	0.054 (-0.028, 0.135)	0.091 (0.006, 0.177)	-0.087 (-0.251, 0.071)
PC1	0.022 (0.002, 0.042)	0.020 (-0.001, 0.041)	0.026 (-0.013, 0.065)
PC2	0.030 (0.00, 0.061)	0.005 (-0.028, 0.038)	0.140 (0.082, 0.198)

Models adjusted for: maternal age, child sex, maternal education, maternal race, parity, pre-pregnancy BMI, marital status, family smoking, and gestational age. Abbreviations: PC1, principal component 1; PC2, principal component 2; PFDS, perfluorodecane sulfonate; PFHxS, perfluorohexanesulfonic acid; PFNA, perfluorononanoic acid; PFOA, perfluorooctanoic acid; PFOS, perfluorooctane sulfonate.

principal component 2 was also significantly associated with higher triglycerides [0.140 (95% CI, 0.082–0.198)].

Discussion

In this study of 222 women who were pregnant during the WTC disaster, the association between cord blood concentrations of PFAS compounds and cord lipid profiles were evaluated. This work was conducted to fill in research gaps on the relationships between cord blood PFAS and lipid profiles and to examine potential health effects resulting from prenatal exposure to the WTC disaster. We found significant associations between greater PFOS, PFOA, and PFHxS with higher total cord lipids; greater PFDS with higher total cholesterol but lower triglycerides; and greater PFOA and PFHxS with higher triglycerides. Quartile and cubic spline analyses were generally consistent and suggested strong linear trends for PFOA and PFHxS with triglycerides. These results build on our previous findings of an association between prenatal WTC exposure and greater PFAS (13) levels by linking prenatal exposure to PFAS with an important and understudied health outcome, cord lipid profiles. Furthermore, they emphasize the need for additional research and continued monitoring of a vulnerable population currently

Table 5. Geometric Mean Ratios of Lipids by PFAS Quartiles (N = 222)

	Total Lipids	Total Cholesterol	Triglycerides
PFOS	1 (Reference)	1 (Reference)	1 (Reference)
Quartile 2	0.96 (0.88 1.04)	0.95 (0.87 1.04)	1.00 (0.84 1.18)
Quartile 3	1.01 (0.93 1.10)	0.99 (0.90 1.08)	1.09 (0.92 1.30)
Quartile 4	1.09 (1.00 1.19)	1.07 (0.97 1.17)	1.16 (0.98 1.38)
	p-trend = 0.03	p-trend = 0.16	p-trend = 0.06
PFOA	1 (Reference)	1 (Reference)	1 (Reference)
Quartile 2	1.04 (0.96 1.13)	1.03 (0.94 1.12)	1.11 (0.94 1.30)
Quartile 3	1.10 (1.01 1.19)	1.09 (1.00 1.20)	1.14 (0.97 1.34)
Quartile 4	1.07 (0.99 1.17)	1.02 (0.93 1.12)	1.33 (1.13 1.57)
	p-trend = 0.04	p-trend = 0.39	p-trend = 0.001
PFNA	1 (Reference)	1 (Reference)	1 (Reference)
Quartile 2	0.99 (0.91 1.08)	0.99 (0.90 1.09)	0.96 (0.81 1.14)
Quartile 3	1.00 (0.91 1.09)	1.00 (0.91 1.10)	0.91 (0.77 1.09)
Quartile 4	1.00 (0.90 1.11)	1.02 (0.91 1.14)	0.87 (0.70 1.07)
	p-trend = 0.94	p-trend = 0.76	p-trend = 0.156
PFHxS	1 (Reference)	1 (Reference)	1 (Reference)
Quartile 2	1.03 (0.94 1.12)	1.03 (0.94 1.13)	1.08 (0.92 1.28)
Quartile 3	1.06 (0.98 1.16)	1.04 (0.95 1.14)	1.22 (1.04 1.45)
Quartile 4	1.07 (0.98 1.16)	1.03 (0.94 1.13)	1.26 (1.07 1.49)
	p-trend = 0.08	p-trend = 0.48	p-trend = 0.002
PFDS	1 (Reference)	1 (Reference)	1 (Reference)
Quartile 2	1.03 (0.95 1.13)	1.03 (0.94 1.13)	1.07 (0.91 1.26)
Quartile 3	0.98 (0.90 1.07)	1.02 (0.93 1.12)	0.77 (0.65 0.91)
Quartile 4	1.06 (0.96 1.17)	1.11 (0.99 1.23)	0.89 (0.74 1.08)
	p-trend = 0.55	p-trend = 0.10	p-trend = 0.04

Models adjusted for: maternal age, child sex, maternal education, maternal race, parity, pre-pregnancy BMI, marital status, family smoking, and gestational age Abbreviations: PFDS, perfluorodecane sulfonate; PFHxS, perfluorohexanesulfonic acid; PFNA, perfluorononanoic acid; PFOA, perfluorooctanoic acid; PFOS, perfluorooctane sulfonate.

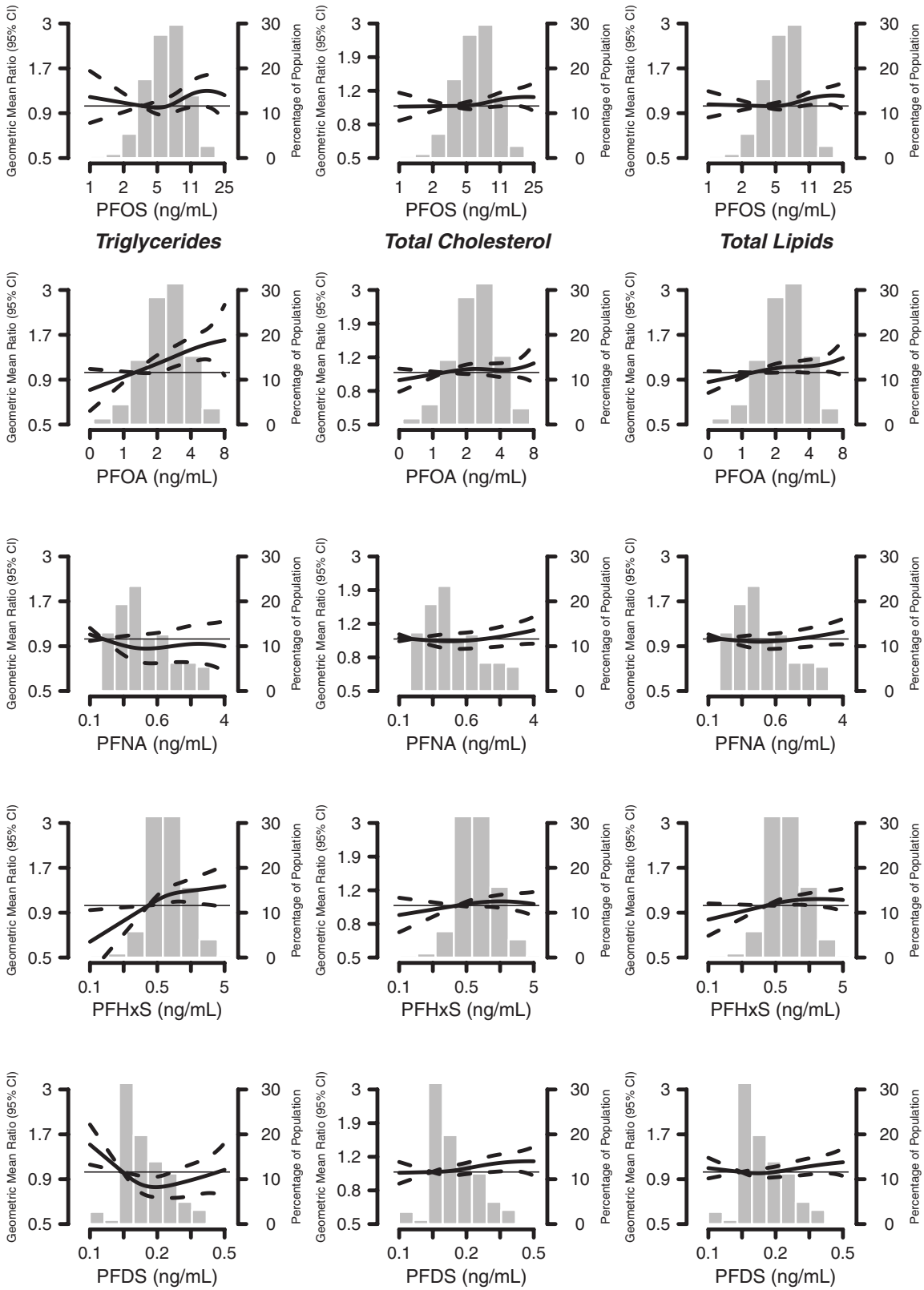

Figure 3. Geometric mean ratio (95% CI) of lipids by PFAS using restricted cubic splines. Lines represent geometric mean ratios of lipids. Shaded areas surrounding the lines represent the 95% confidence intervals. The reference was set at the 10th percentile of each PFAS distribution. Geometric mean ratios were adjusted for maternal age, child sex, maternal education, maternal race, parity, pre-pregnancy BMI, marital status, family smoking, and gestational age. Abbreviations: PFDS, perfluorodecane sulfonate; PFHxS, perfluorohexanesulfonic acid; PFNA, perfluorononanoic acid; PFOA, perfluorooctanoic acid; PFOS, perfluorooctane sulfonate.

entering adulthood who were prenatally exposed to the WTC disaster.

Cardiovascular disease is the leading cause of death both nationally (14) and globally (15), contributing to more than 17 million deaths worldwide (42). Evidence suggests that environmental factors play a significant role in the development and severity of the disease (43); given their ubiquity and persistence, there has been growing interest in the contribution of PFAS to environmental risk for cardiovascular disease. Epidemiologic studies have identified significant associations between PFAS exposure and numerous cardiometabolic outcomes (19, 20, 24, 44, 45). Several studies have also reported on the association between PFAS compounds and lipid profiles in adults (19–21, 25), children (4, 17, 18, 22, 23), occupationally exposed groups (37, 46–50), and pregnant women (26, 27). Dyslipidemias, including elevated total serum cholesterol (51–53)and elevated triglycerides (54), are well-established risk factors for development of cardiovascular disease. Findings generally suggest a positive trend between PFAS and both triglycerides and total cholesterol; however, the significance of relationships varies by compound across studies. Furthermore, there have been some inconsistent results, including negative associations between PFHxS and total cholesterol (20), as well as between triglycerides and PFNA and PFOS (27).

Understanding of the biological mechanism behind the association between PFAS and lipids is limited. Human genetic studies have found PFOA and PFOS to be associated with differential expression of genes related to lipid transport and metabolism, which may explain the positive relationship reported between PFAS and cholesterol (55). However, PFAS are known to activate the peroxisome proliferator-activated alpha receptor (PPARα) (56), a regulator of lipid homeostasis, which results in reductions of cholesterol and triglycerides in serum and an accumulation of lipids in the liver (57). This mechanism is consistent with the hypolipidemic effect of PFAS observed in animal studies (57–59), but does not explain the positive relationship observed in epidemiologic studies. Peroxisome proliferation is more apparent in animals (60), so it is possible that this mechanism is less important in humans, highlighting the need for additional research to better understand the mode of action behind this relationship.

PFAS have been shown to cross the placenta in humans (11, 12). In turn, several prospective birth cohorts have also attempted to understand the association between prenatal exposure to PFAS and birth outcomes, as well as health effects in childhood. Stemming from the Barker Hypothesis nearly 3 decades ago, there has been a growing emphasis on fetal origins of cardiometabolic outcomes later in life (61). Indeed, some evidence suggests that cord lipid profiles may predict future lipid levels (62). Furthermore, higher cord lipid levels have been consistently reported in low-birth-weight and small-for-gestational-age newborns, both established risk factors for cardiovascular disease in adulthood (63, 64). Few studies have evaluated the association between prenatal exposure and effects on lipid profiles, all of which report the relationship between maternal exposure during pregnancy and subsequent effects on lipid levels in childhood (28–30). Most findings have been null, however positive associations were observed between PFHxS and triglycerides (29), as well as between PFOA and total cholesterol (30). To our knowledge, no studies have evaluated the relationship between PFAS exposure and cord lipid profiles. Measuring both PFAS and lipids in cord blood may reflect the exposure-outcome relationship more accurately; studies have suggested PFAS vary in their transplacental transfer efficiency, affecting the correlation between maternal and cord blood PFAS (31–33). In our study, we observed several significant associations between cord PFAS and cord lipids, including evidence of a dose-response relationship for both PFOA and PFHxS with triglycerides. Of note, the association between PFOA and triglycerides is consistent with findings from a cross-sectional analysis of serum PFAS and lipids measured in adolescents enrolled in the World Trade Center Health Registry (children exposed to the WTC disaster either prenatally or during childhood) and a matched comparison group (17). We also identified a novel association between PFDS (a PFAS compound with limited available toxicological information) and total cholesterol. This observed relationship, despite low cord levels of PFDS in comparison to the other PFAS measured, highlights the need for more research on this compound as well as other understudied PFAS that may be present at lower quantities than the more well-known PFAS. Interestingly, we also observed a negative association between PFDS and triglycerides in quartile analyses, again, emphasizing the need for more research to understand the mechanisms behind this PFAS and its association with lipids.

Cord PFOA, PFNA, and PFHxS levels in our population were similar to 2 other US studies (65, 66) that have evaluated cord PFAS, suggesting these findings are relevant to the exposures seen in the general population. Cord PFOS levels, however, were slightly higher in our population (geometric mean = 6.29 ng/mL) than concentrations in both Baltimore, MD (65) and Cincinnati, OH (66) (geometric means = 1.6 and 3.32 ng/mL, respectively) birth cohorts. To our knowledge, cord PFDS concentrations have not been reported in any other birth cohorts, emphasizing the novelty of these findings.

In conclusion, we found significant associations between several cord blood PFAS and cord lipids, presenting novel findings in cord blood, but consistent with the mounting evidence of a relationship between PFAS and adverse cardiometabolic outcomes, even at low levels of exposure. Furthermore, in a previous study conducted in this population, we found a significant association between living or working within 2 miles of the WTC disaster and prenatal exposure to PFOA, identifying this group as a potentially vulnerable population and emphasizing the importance of additional studies evaluating health effects arising from this exposure. Our associations between cord PFAS and altered cord lipid profiles in this study, including with PFOA and both triglycerides and total lipids, provides one potential pathway between prenatal WTC exposure and adverse health outcomes. Continued monitoring of this population and additional studies are needed to evaluate whether these exposures are associated with health effects as the population ages into adulthood.

Acknowledgments

The authors wish to thank Dr Andreas Sjodin from the Centers for Disease Control and Prevention for his work on lipid measurements.

***Financial Support*:** This work was supported by grants 1U01-OH010714-01A1, 1U01-OH010394-01A1, 1U01-OH011299-01A1 from NIOSH; and the National Institute of Environmental Health Sciences grant ES09089.

Additional Information

Correspondence: Miranda J. Spratlen, Department of Environmental Health Sciences, Columbia University, 122 W 168th, Room 1105, New York, NY 10032. E-mail: mjs2376@cumc.columbia.edu.

Disclosure Summary: The authors have nothing to disclose.

References

1. United States Environmental Protection Agency (EPA). *Technical Fact Sheet – Perfluorooctane Sulfonate (PFOS) and Perfluorooctanoic Acid (PFOA). EPA 505-F-17-001*. Washington, DC; 2017. https://www.epa.gov/sites/production/files/2017-12/documents/ffrrofactsheet_contaminants_pfos_pfoa_11-20-17_508_0.pdf
2. Trudel D, Horowitz L, Wormuth M, Scheringer M, Cousins IT, Hungerbuhler K. Estimating consumer exposure to PFOS and PFOA. *Risk Anal*. 2008;**28**(2):251–269.
3. Fraser AJ, Webster TF, Watkins DJ, Nelson JW, Stapleton HM, Calafat AM, Kato K, Shoeib M, Vieira VM, McClean MD. Polyfluorinated compounds in serum linked to indoor air in office environments. *Environ Sci Technol*. 2012;**46**(2):1209–1215.
4. Jain RB, Ducatman A. Associations between lipid/lipoprotein levels and perfluoroalkyl substances among US children aged 6–11 years. *Environ Pollut*. 2018;**243**(Pt A):1–8.
5. Kato K, Wong LY, Jia LT, Kuklenyik Z, Calafat AM. Trends in exposure to polyfluoroalkyl chemicals in the U.S. Population: 1999–2008. *Environ Sci Technol*. 2011;**45**(19):8037–8045.
6. Agency EP. Lifetime health advisories and health effects support documents for perfluorooctanoic acid and perfluorooctane sulfonate. *Federal Register*. 2016;**81**(101):33250–33251.
7. *Commission Recommendation 2010/161/EU of 17 March 2010 on the monitoring of perfluoroalkylated substances in food*. Official Journal of the European Union; 2010:22–23.
8. *Proposal for a directive of the European Parliament and of the Council on the quality of water intended for human consumption (recast). COM/2017/0753 final – 2017/0332 (COD)*. European Commission. Brussels, Belgium; 2018.
9. EPA. *Health Effects Support Document for Perfluorooctanoic Acid (PFOA). EPA 822-R-16-003*. Washington, DC; 2016. https://www.epa.gov/sites/production/files/2016-05/documents/pfoa_hesd_final-plain.pdf
10. EPA. *Health Effects Support Document for Perfluorooctane Sulfonate (PFOS)*. EPA 822-R-16-002. Washington, DC; 2016. https://www.epa.gov/sites/production/files/2016-05/documents/pfos_hesd_final_508.pdf
11. Olsen GW, Butenhoff JL, Zobel LR. Perfluoroalkyl chemicals and human fetal development: an epidemiologic review with clinical and toxicological perspectives. *Reprod Toxicol*. 2009;**27**(3–4):212–230.
12. Bach CC, Bech BH, Brix N, Nohr EA, Bonde JP, Henriksen TB. Perfluoroalkyl and polyfluoroalkyl substances and human fetal growth: a systematic review. *Crit Rev Toxicol*. 2015;**45**(1):53–67.
13. Spratlen MJ, Perera FP, Lederman SA, Robinson M, Kannan K, Trasande L, Herbstman J. Cord blood perfluoroalkyl substances in mothers exposed to the World Trade Center disaster during pregnancy. *Environ Pollut*. 2019;**246**:482–490.
14. *Heart Disease Fact Sheet*. August 23, 2017. 2017.
15. *Global Health Estimates 2016: Deaths by Cause, Age, Sex, by Country and by Region, 2000–2016*. Geneva, Switzerland: World Health Organization; 2018.
16. Yeung EH, Robledo C, Boghossian N, Zhang C, Mendola P. Developmental origins of cardiovascular disease. *Curr Epidemiol Rep*. 2014;**1**(1):9–16.
17. Koshy TT, Attina TM, Ghassabian A, Gilbert J, Burdine LK, Marmor M, Honda M, Chu DB, Han X, Shao Y, Kannan K, Urbina EM, Trasande L. Serum perfluoroalkyl substances and cardiometabolic consequences in adolescents exposed to the World Trade Center disaster and a matched comparison group. *Environ Int*. 2017;**109**:128–135.
18. Zeng XW, Qian Z, Emo B, Vaughn M, Bao J, Qin XD, Zhu Y, Li J, Lee YL, Dong GH. Association of polyfluoroalkyl chemical exposure with serum lipids in children. *Sci Total Environ*. 2015;**512–513**:364–370.
19. Christensen KY, Raymond M, Meiman J. Perfluoroalkyl substances and metabolic syndrome. *Int J Hyg Environ Health*. 2019;**222**(1):147–153.
20. Nelson JW, Hatch EE, Webster TF. Exposure to polyfluoroalkyl chemicals and cholesterol, body weight, and insulin resistance in the general U.S. population. *Environ Health Perspect*. 2010;**118**(2):197–202.
21. Fisher M, Arbuckle TE, Wade M, Haines DA. Do perfluoroalkyl substances affect metabolic function and plasma lipids?--Analysis of the 2007–2009, Canadian Health Measures Survey (CHMS) Cycle 1. *Environ Res*. 2013;**121**:95–103.
22. Frisbee SJ, Shankar A, Knox SS, Steenland K, Savitz DA, Fletcher T, Ducatman AM. Perfluorooctanoic acid, perfluorooctanesulfonate, and serum lipids in children and adolescents: results from the C8 Health Project. *Arch Pediatr Adolesc Med*. 2010;**164**(9):860–869.

23. Geiger SD, Xiao J, Ducatman A, Frisbee S, Innes K, Shankar A. The association between PFOA, PFOS and serum lipid levels in adolescents. *Chemosphere*. 2014;**98**:78–83.
24. Liu HS, Wen LL, Chu PL, Lin CY. Association among total serum isomers of perfluorinated chemicals, glucose homeostasis, lipid profiles, serum protein and metabolic syndrome in adults: NHANES, 2013–2014. *Environ Pollut*. 2018;**232**:73–79.
25. Eriksen KT, Raaschou-Nielsen O, McLaughlin JK, Lipworth L, Tjonneland A, Overvad K, Sorensen M. Association between plasma PFOA and PFOS levels and total cholesterol in a middle-aged Danish population. Published February 18, 2013. *PLoS One*. 2013;**8**(2):e56969. doi:10.1371/journal.pone.0056969.
26. Starling AP, Engel SM, Whitworth KW, Richardson DB, Stuebe AM, Daniels JL, Haug LS, Eggesbo M, Becher G, Sabaredzovic A, Thomsen C, Wilson RE, Travlos GS, Hoppin JA, Baird DD, Longnecker MP. Perfluoroalkyl substances and lipid concentrations in plasma during pregnancy among women in the Norwegian Mother and Child Cohort Study. *Environ Int*. 2014;**62**:104–112.
27. Matilla-Santander N, Valvi D, Lopez-Espinosa MJ, Manzano-Salgado CB, Ballester F, Ibarluzea J, Santa-Marina L, Schettgen T, Guxens M, Sunyer J, Vrijheid M. Exposure to perfluoroalkyl substances and metabolic outcomes in pregnant women: evidence from the Spanish INMA birth cohorts. *Environ Health Perspect*. 2017;**125**(11):117004. doi:10.1289/EHP1062
28. Mora AM, Fleisch AF, Rifas-Shiman SL, Woo Baidal JA, Pardo L, Webster TF, Calafat AM, Ye X, Oken E, Sagiv SK. Early life exposure to per- and polyfluoroalkyl substances and mid-childhood lipid and alanine aminotransferase levels. *Environ Int*. 2018;**111**:1–13.
29. Manzano-Salgado CB, Casas M, Lopez-Espinosa MJ, Ballester F, Iniguez C, Martinez D, Romaguera D, Fernandez-Barres S, Santa-Marina L, Basterretxea M, Schettgen T, Valvi D, Vioque J, Sunyer J, Vrijheid M. Prenatal exposure to perfluoroalkyl substances and cardiometabolic risk in children from the Spanish INMA birth cohort study. *Environ Health Perspect*. 2017;**125**(9):097018.
30. Maisonet M, Nayha S, Lawlor DA, Marcus M. Prenatal exposures to perfluoroalkyl acids and serum lipids at ages 7 and 15 in females. *Environ Int*. 2015;**82**:49–60.
31. Midasch O, Drexler H, Hart N, Beckmann MW, Angerer J. Transplacental exposure of neonates to perfluorooctanesulfonate and perfluorooctanoate: a pilot study. *Int Arch Occup Environ Health*. 2007;**80**(7):643–648.
32. Winkens K VR, Berger U, Cousins I. Early life exposure to per- and polyfluoroalkyl substances (PFASs): a critical review. *Emerging Contaminants*. 2017;**3**(2):55–68.
33. Fei C, McLaughlin JK, Tarone RE, Olsen J. Perfluorinated chemicals and fetal growth: a study within the Danish National Birth Cohort. *Environ Health Perspect*. 2007;**115**(11):1677–1682.
34. Lederman SA, Rauh V, Weiss L, Stein JL, Hoepner LA, Becker M, Perera FP. The effects of the World Trade Center event on birth outcomes among term deliveries at three lower Manhattan hospitals. *Environ Health Perspect*. 2004;**112**(17):1772–1778.
35. Kannan K, Corsolini S, Falandysz J, Fillmann G, Kumar KS, Loganathan BG, Mohd MA, Olivero J, Van Wouwe N, Yang JH, Aldoust KM. Perfluorooctanesulfonate and related fluorochemicals in human blood from several countries. *Environ Sci Technol*. 2004;**38**(17):4489–4495.
36. Taniyasu S, Kannan K, So MK, Gulkowska A, Sinclair E, Okazawa T, Yamashita N. Analysis of fluorotelomer alcohols, fluorotelomer acids, and short- and long-chain perfluorinated acids in water and biota. *J Chromatogr A*. 2005;**1093**(1–2):89–97.
37. Sakr CJ, Kreckmann KH, Green JW, Gillies PJ, Reynolds JL, Leonard RC. Cross-sectional study of lipids and liver enzymes related to a serum biomarker of exposure (ammonium perfluorooctanoate or APFO) as part of a general health survey in a cohort of occupationally exposed workers. *J Occup Environ Med*. 2007;**49**(10):1086–1096.
38. Kataria A, Trachtman H, Malaga-Dieguez L, Trasande L. Association between perfluoroalkyl acids and kidney function in a cross-sectional study of adolescents. *Environ Health*. 2015;**14**:89.
39. Phillips DL, Pirkle JL, Burse VW, Bernert Jr JT, Henderson LO, Needham LL. Chlorinated hydrocarbon levels in human serum: effects of fasting and feeding. *Arch Environ Contam Toxicol*. 1989;**18**(4):495–500.
40. Hoffman K, Webster TF, Weisskopf MG, Weinberg J, Vieira VM. Exposure to polyfluoroalkyl chemicals and attention deficit/hyperactivity disorder in U.S. children 12–15 years of age. *Environ Health Perspect*. 2010;**118**(12):1762–1767.
41. Kaiser H. The varimax criterion for analytic rotation in factor analysis. *Psychometrika*. 1958;**23**:187–200.
42. *Cardiovascular diseases (CVDs)*. http://www.who.int/en/news-room/fact-sheets/detail/cardiovascular-diseases-(cvds). Accessed December 5, 2018.
43. Bhatnagar A. Environmental determinants of cardiovascular disease. *Circ Res*. 2017;**121**(2):162–180.
44. Cardenas A, Gold DR, Hauser R, Kleinman KP, Hivert MF, Calafat AM, Ye X, Webster TF, Horton ES, Oken E. Plasma concentrations of per- and polyfluoroalkyl substances at baseline and associations with glycemic indicators and diabetes incidence among high-risk adults in the diabetes prevention program trial. *Environ Health Perspect*. 2017;**125**(10):107001. doi:10.1289/EHP1612
45. Shankar A, Xiao J, Ducatman A. Perfluorooctanoic acid and cardiovascular disease in US adults. *Arch Intern Med*. 2012;**172**(18):1397–1403.
46. Steenland K, Tinker S, Frisbee S, Ducatman A, Vaccarino V. Association of perfluorooctanoic acid and perfluorooctane sulfonate with serum lipids among adults living near a chemical plant. *Am J Epidemiol*. 2009;**170**(10):1268–1278.
47. Costa G, Sartori S, Consonni D. Thirty years of medical surveillance in perfluorooctanoic acid production workers. *J Occup Environ Med*. 2009;**51**(3):364–372.
48. Sakr CJ, Leonard RC, Kreckmann KH, Slade MD, Cullen MR. Longitudinal study of serum lipids and liver enzymes in workers with occupational exposure to ammonium perfluorooctanoate. *J Occup Environ Med*. 2007;**49**(8):872–879.
49. Olsen GW, Burris JM, Burlew MM, Mandel JH. Plasma cholecystokinin and hepatic enzymes, cholesterol and lipoproteins in ammonium perfluorooctanoate production workers. *Drug Chem Toxicol*. 2000;**23**(4):603–620.
50. Olsen GW, Zobel LR. Assessment of lipid, hepatic, and thyroid parameters with serum perfluorooctanoate (PFOA) concentrations in fluorochemical production workers. *Int Arch Occup Environ Health*. 2007;**81**(2):231–246.
51. Blesso CN, Fernandez ML. Dietary cholesterol, serum lipids, and heart disease: are eggs working for or against you? *Nutrients*. 2018;**10**(4):E426. doi:10.3390/nu10040426
52. Nagasawa SY, Okamura T, Iso H, Tamakoshi A, Yamada M, Watanabe M, Murakami Y, Miura K, Ueshima H, Evidence for cardiovascular prevention from observational cohorts in Japan Research G. Relation between serum total cholesterol level and cardiovascular disease stratified by sex and age group: a pooled analysis of 65 594 individuals from 10 cohort studies in Japan. *J Am Heart Assoc*. 2012;**1**(5):e001974. doi:10.1161/JAHA.112.001974
53. Prospective Studies C, Lewington S, Whitlock G, Clarke R, Sherliker P, Emberson J, Halsey J, Qizilbash N, Peto R, Collins R. Blood cholesterol and vascular mortality by age, sex, and blood pressure: a meta-analysis of individual data from 61 prospective studies with 55,000 vascular deaths. *Lancet*. 2007;**370**(9602):1829–1839.
54. Nordestgaard BG, Varbo A. Triglycerides and cardiovascular disease. *Lancet*. 2014;**384**(9943):626–635.
55. Fletcher T, Galloway TS, Melzer D, Holcroft P, Cipelli R, Pilling LC, Mondal D, Luster M, Harries LW. Associations between PFOA,

PFOS and changes in the expression of genes involved in cholesterol metabolism in humans. *Environ Int*. 2013;**57–58**:2–10.
56. Wolf CJ, Takacs ML, Schmid JE, Lau C, Abbott BD. Activation of mouse and human peroxisome proliferator-activated receptor alpha by perfluoroalkyl acids of different functional groups and chain lengths. *Toxicol Sci*. 2008;**106**(1):162–171.
57. Kennedy GL, Jr., Butenhoff JL, Olsen GW, O'Connor JC, Seacat AM, Perkins RG, Biegel LB, Murphy SR, Farrar DG. The toxicology of perfluorooctanoate. *Crit Rev Toxicol*. 2004;**34**(4):351–384.
58. Lau C, Anitole K, Hodes C, Lai D, Pfahles-Hutchens A, Seed J. Perfluoroalkyl acids: a review of monitoring and toxicological findings. *Toxicol Sci*. 2007;**99**(2):366–394.
59. Loveless SE, Finlay C, Everds NE, Frame SR, Gillies PJ, O'Connor JC, Powley CR, Kennedy GL. Comparative responses of rats and mice exposed to linear/branched, linear, or branched ammonium perfluorooctanoate (APFO). *Toxicology*. 2006;**220**(2–3):203–217.
60. DeWitt JC, Shnyra A, Badr MZ, Loveless SE, Hoban D, Frame SR, Cunard R, Anderson SE, Meade BJ, Peden-Adams MM, Luebke RW, Luster MI. Immunotoxicity of perfluorooctanoic acid and perfluorooctane sulfonate and the role of peroxisome proliferator-activated receptor alpha. *Crit Rev Toxicol*. 2009;**39**(1):76–94.
61. Barker DJ. Fetal origins of coronary heart disease. *Br Heart J*. 1993;**69**(3):195–196.
62. Fonnebo V, Dahl LB, Moe PJ, Ingebretsen OC. Does VLDL-LDL-cholesterol in cord serum predict future level of lipoproteins? *Acta Paediatr Scand*. 1991;**80**(8–9):780–785.
63. Smith CJ, Ryckman KK, Barnabei VM, Howard BV, Isasi CR, Sarto GE, Tom SE, Van Horn LV, Wallace RB, Robinson JG. The impact of birth weight on cardiovascular disease risk in the Women's Health Initiative. *Nutr Metab Cardiovasc Dis*. 2016;**26**(3):239–245.
64. Crispi F, Miranda J, Gratacos E. Long-term cardiovascular consequences of fetal growth restriction: biology, clinical implications, and opportunities for prevention of adult disease. *Am J Obstet Gynecol*. 2018;**218**(2S):S869–S879.
65. Apelberg BJ, Goldman LR, Calafat AM, Herbstman JB, Kuklenyik Z, Heidler J, Needham LL, Halden RU, Witter FR. Determinants of fetal exposure to polyfluoroalkyl compounds in Baltimore, Maryland. *Environ Sci Technol*. 2007;**41**(11):3891–3897.
66. Kato K, Wong LY, Chen A, Dunbar C, Webster GM, Lanphear BP, Calafat AM. Changes in serum concentrations of maternal poly- and perfluoroalkyl substances over the course of pregnancy and predictors of exposure in a multiethnic cohort of Cincinnati, Ohio pregnant women during 2003–2006. *Environ Sci Technol*. 2014;**48**(16):9600–9608.