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Prenatal Exposure to Perfluoroalkyl Substances and Body Fatness in Girls

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Abstract

Background: Perfluoroalkyl substances (PFASs) are used in surface coatings that resist stains, grease, and water.

Methods: The association between *in utero* PFAS exposure and girls' body fatness at age 9 was analyzed in The Avon Longitudinal Study of Parents and Children (UK). Maternal serum [median 15 weeks: interquartile range (IQR) 10 and 28 weeks of gestation] was analyzed for perfluorooctane sulfonate (PFOS), perfluorooctanoate (PFOA), perfluorohexane sulfonate (PFHxS), and perfluorononanoate (PFNA). Body composition was measured by dual X-ray emission absorptiometry, and percent total body fat (%BF) was calculated. Associations between PFASs and body fatness were modeled by multivariable linear regression.

Results: Among 359 girls, median (IQR) %BF was 27.5 (IQR 21.7–34.6). Median (IQR) concentrations (all ng/mL) were 3.7 (2.9–4.8) for PFOA, 19.8 (15.0–25.3) for PFOS, 1.6 (1.3–2.2) for PFHxS, and 0.5 (0.4–0.7) for PFNA. Maternal PFAS concentrations were not significantly associated with daughters' total %BF overall. Mothers' educational status modified associations for PFOA and PFOS with %BF (P-interactions: 0.005 and 0.02, respectively). %BF was higher [1.4%; 95% confidence interval (95% CI): 0.3 to 2.5] for each one unit (ng/mL) higher PFOA among girls with mothers in the middle education group, but lower (−0.6%; 95% CI: −1.12 to −0.04) for the corresponding comparison among girls with mothers with the highest education. %BF was lower (−0.2%; 95% CI: −0.3 to −0.1) for each one unit higher PFOS among girls with the most educated mothers.

Conclusions: Prenatal exposure to PFOA and PFOS was associated with girls' %BF within some strata of maternal education status. PFHxS and PFNA were not associated with %BF.

Keywords: body composition; epidemiology; obesity

Introduction

In 2012, the prevalence of overweight and obesity among girls aged 2–10 years in the United Kingdom was estimated at 22.8%.[1,2] Increased dietary energy intakes and sedentary behaviors conducive to positive energy balance are well-known risk factors for obesity. Emerging evidence suggests that there may also be mechanisms through which early life (*e.g.*, *in utero*, infancy) exposure to a subclass of environmental endocrine-disrupting chemicals (EDCs) may contribute to altered growth and development with long-term effects on recognized risk factors for chronic disease.[3–5] For example, certain EDCs could disrupt hormonally regulated ingestive behaviors or metabolic sequelae resulting in a metabolic phenotype with a predisposition to gain weight.[4] Excess weight during childhood is associated with adverse effects on circulating blood lipids, measures of insulin resistance, and elevated blood pressure known to persist into adulthood and predict adult cardiometabolic risk factors and chronic disease.[6–9]

Perfluoroalkyl substances (PFASs) are synthetically made chemicals used as surfactants and surface coatings to decrease staining and sticking.[10] PFASs are used commercially as lubricants, paper and textile coatings, in polishes, in aqueous film-forming foams (fire fighting), and in

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food packaging materials.[10] Exposure to PFASs is widespread; detectable levels were found in more than 98% of Americans who participated in the US National Health and Nutrition Examination Survey (NHANES) from 2003 to 2004.[11] Because of their persistence and tendency to bioaccumulate, there is growing interest in the relationship between exposure, especially during critical windows of susceptibility such as early life, and health effects later on.[12] Exposure to PFASs during development could permanently influence risk for chronic disease later in life through long-term effects on risk factors such as weight gain and body fatness.[3]

Prenatal exposure to PFASs has been linked with smaller size at birth.[13–15] In follow-up analyses of girls at 20 months of age, those with higher prenatal serum concentrations of perfluorooctane sulfonate (PFOS) were heavier despite being smaller at birth.[12] The goal of this research was to assess the role of prenatal exposure to PFASs on adiposity measured in older girls. Our analyses used existing prospectively collected data from the Avon Longitudinal Study of Parents and Children (ALSPAC) to evaluate the association between maternal serum concentrations measured during pregnancy of PFASs and body fatness measured by dual-energy X-ray absorptiometry (DXA) in girls at age 9 years.

Methods

Participants

The ALSPAC is a prospective birth cohort in the United Kingdom that enrolled 14,541 pregnant women residing in Avon, UK, with expected delivery dates between April 1991 and December 1992.[16,17] The goal of ALSPAC was to study the effects of genetics, lifestyle factors, and the physical environment on the health, behavior, and development of children. This study includes a subset of mother–daughter dyads from the parent study selected for an ancillary study of maternal serum concentrations of environmental exposures and daughter's puberty characteristics.[18] To be considered, girls had to have at least two pubertal assessments to allow for classification of age at menarche. The ancillary study included all girls with early menarche (<11.5 years; $N=218$) and a random sample of girls without early menarche (≥11.5 years; $N=230$). Informed consent was provided at the time of enrollment by the mothers. Human subjects' protection and ethical approval was provided by the ALSPAC Law and Ethics Committee, the Local Research Ethics Committees, and the CDC Institutional Review Board.

Data Collection

Mothers reported demographic (*e.g.*, age, educational status, race/ethnicity), health (*e.g.*, prepregnancy BMI kg/m^2), and lifestyle (*e.g.*, smoking status) information for themselves at enrollment and later on annually for their children. Birth characteristics, including weight (grams), length (centimeters), and gestational age (weeks), were abstracted from medical records. Since enrollment, detailed information has been collected on the children using parent or guardian- and self-reported questionnaires that address growth and development, psychological, social, and health behaviors, and a variety of health-related outcomes.

Trained research assistants reviewed these records with mothers as they were completed. In addition, periodic clinical assessments have collected detailed physiological data, cognitive information, and biological samples.[16,19,20] Measures of total and regional body fat, lean mass, and bone mass were made biennially using a Lunar Prodigy DXA scanner (GE Medical Systems Lunar, Madison, WI) beginning at age 9 years. Height was measured without shoes to the closest millimeter with a Harpenden stadiometer (Holtain Ltd., Crosswell, UK) and weight was measured using a body fat analyzer (model TBF 305; Tanita UK Ltd., Yiewsley, UK). BMI was calculated as weight (kg)/height (m^2). Waist circumference (WC) was assessed with a tape measure to the closest millimeter (Holtain).

The main outcome variables we used were percent total body fat (%BF) and as a secondary measure, the ratio of trunk fat mass to total fat mass [(trunk fat (kg)/total fat (kg)) *100] (%TF). Gynoid fat mass was not measured at age 9; thus, we could not calculate the android:gynoid fat mass ratio. The coefficient of variations for total body fat and truncal fat in this population was reported previously as 2.3% and 6.2%, respectively.[21]

We conducted analyses for BMI and WC as sensitivity analyses and to allow for comparisons with reports from other research. At age 9, a total of 359 of 448 girls whose mothers had previously provided prenatal sera for PFAS measurements also contributed DXA scans for assessment of body fatness. The ALSPAC website contains details of all the data that are available through a fully searchable data dictionary available at www.bris.ac.uk/alspac/researchers/data-access/data-dictionary.

Laboratory Analyses

At enrollment (1991–1992), mothers were asked to provide a single pregnancy blood sample as a measure of prenatal exposure, which was processed, frozen, and stored for later analysis. Median gestational age at collection was 15 weeks with an interquartile range (IQR) of 10–28 weeks (Supplementary Table S1; Supplementary Data are available at www.liebertpub.com/chi). Perfluorooctanoate (PFOA), PFOS, perfluorohexane sulfonate (PFHxS), and perfluorononanoate (PFNA) were measured in stored maternal serum samples from mothers of girls by the National Center for Environmental Health laboratories of the CDC (Atlanta, GA) as described elsewhere.[12] Briefly, sera samples were analyzed by online solid-phase extraction coupled to isotope dilution high-performance liquid chromatography/tandem mass spectrometry. Analytes were detected in 100% of samples. The limit of detection for PFNA was 0.08 ng/mL, for PFOA and PFHxS was 0.10 ng/mL, and for PFOS was 0.20 ng/mL. Laboratory analyses included low-concentration and high-concentration pooled standards,

reagent blanks, and study samples. Precision of the measurements for the four PFASs was 8%–13%.

Statistical Analyses

The laboratory data were plotted and examined for outliers. Measures of central tendency and distributions were calculated for demographic characteristics of the mother–daughter dyads. Spearman correlation coefficients were calculated between the PFASs. Our analyses were conducted on a sample of girls previously selected for a nested case–control study to evaluate the association of PFASs with early age at menarche. PFAS concentrations did not appear to be associated with age at menarche in this sample[18]; however, to account for the sampling selection probabilities, we conservatively constructed stratum-weighted linear regression models to account for the sampling scheme used for participant selection,[22] weighting girls who attained menarche <11.5 years at 1 and the girls who attained menarche at 11.5 years or older 15.1 (a random sample of the girls who attained menarche ≥11.5 years of age) as described previously.[18]

These regression models were used to evaluate the associations of girls' total body fatness (%BF) and trunk:total fat mass (%TF) at 9 years of age, with each PFAS after adjustment for maternal prepregnancy BMI (continuous) and educational status (low, medium, and high). To evaluate the potential for nonlinearity, we conducted residual analyses plotting the residuals vs. independent variables and assessed the square root and squared terms of PFASs in models as continuous variables. There was no evidence of nonlinearity for any of the associations between PFASs and body fatness variables.

In this analysis, not attaining any General Certificates of Secondary Education (GCSEs, at 16 years of age) was coded as ''low'' educational status, obtaining GCSEs as ''medium,'' and completing GCSEs with additional education (*e.g.*, university) was considered ''high.'' Smoking was an important confounder only in the PFNA models in our analyses; thus, PFNA models were also adjusted for confounding by maternal smoking status (yes/no any prenatal). A number of other potential confounders were considered, including maternal age (continuous), race (Caucasian vs. other), parity (continuous), gestational age at blood collection (continuous), previous live birth (yes/no), daughters' preterm delivery (<37 weeks yes/no), birthweight (continuous, gm), breastfed (any vs. none), activity level in childhood (very active vs. not), exact age at DXA scan, and girls dietary energy and macronutrient intake at age 7 (continuous total energy, percent energy from fat, protein, and carbohydrate). None of these additional variables improved model fit or led to meaningful changes in the relationship between body fatness measures and PFASs (modified β estimates >10%); thus, they were not retained in the final models.

We considered potential effect modification by maternal prepregnancy BMI, smoking, and educational status and by daughters' birthweight, by including these variables and their cross-product terms with each of the PFASs in their respective models and by using the likelihood ratio test to compare hierarchically nested models. Finally, in sensitivity analyses and to allow for comparison with other research, we evaluated the associations between maternal PFASs with daughters' BMI and WC. Statistical analyses were performed using Statistical Analysis Systems (SAS version 9.3) software.

Results

Table 1 presents PFAS levels by sample demographic characteristics. Overall, the sample was primarily Caucasian (>90% of mothers and girls). The majority (64%) of mothers were considered normal weight before pregnancy and nearly half of mothers reported a previous live birth. There were few girls who were preterm or of low birthweight. Of the PFASs (all ng/mL) measured, concentrations of PFOS were the highest (median 19.7; IQR: 15.0–25.3) and PFNA concentrations were the lowest (median 0.5; IQR: 0.4–0.7). Spearman correlation coefficients between the PFASs ranged between 0.25 for PFNA and PFHxS to 0.71 for PFOA and PFOS (data not presented). Median concentrations of PFOS and PFOA tended to be higher among mothers who were more educated and nonsmokers. All the measured PFASs tended to be higher among mothers who had reported not having a previous live birth or who reported a birth that was preterm or of low birthweight (Table 1). Among daughters, median %BF was 27.5 (IQR 21.7–34.6), %TF was 39.6 (IQR 36.5–43.7), BMI (kg/m^2) was 18.1 (IQR 16.3–20.6), and WC (cm) was 62.7 (IQR 58.6–68.8) (Supplementary Table S1).

In multivariable regression analyses (Table 2), mothers' PFASs were not associated with daughters' %BF overall (main effects). Mothers' prepregnancy BMI and smoking status and daughters' birthweight did not significantly modify the associations between maternal PFAS concentrations and daughters' %BF.

We observed modification by mothers' educational status for PFOA and PFOS (P-interactions: 0.005 and 0.02, respectively). To be consistent, for all PFASs we present education-stratified results that allow for a different slope across the three educational categories (P-interaction >0.05 is not considered significant). (Supplementary Tables S2 and S3 and Figures S1 and S2 are also available.) For PFOA, %BF was significantly higher (1.4%; 95% confidence interval [95% CI]: 0.3 to 2.5) for each one unit (ng/mL) higher PFOA among girls with mothers in the middle education group ($\sim$5% difference from the median), but slightly lower (–0.6%; 95% CI: −1.12 to −0.04) for the corresponding comparison among girls with mothers in the highest education group. For PFOS, %BF was significantly lower (–0.2%; 95% CI: −0.3 to −0.1) for each one unit higher PFOS only among girls with mothers in the highest education group.

For the outcome %TF, we observed modification by mothers' educational status for PFOS and PFHxS (P-interactions: 0.006 and 0.04, respectively). For PFOS, the pattern for %TF was similar to that observed with total %BF, but for PFHxS, although the interaction was

Table 1. Characteristics of Study Population (*N* = 359)

	Frequency *N* (%)	PFOS (ng/mL)	PFOA (ng/mL)	PFHxS (ng/mL)	PFNA (ng/mL)
		Median (IQR)			
Overall	359 (100)	19.7 (15.0–25.3)	3.7 (2.9–4.8)	1.6 (1.3–2.2)	0.5 (0.4–0.7)
Maternal prepregnancy BMI					
Underweight (<18.5)	16 (4.5)	16.9 (13.2–23.7)	3.7 (2.8–4.8)	1.5 (1.2–2.3)	0.5 (0.3–0.6)
Normal (18.5–24.9)	229 (63.8)	20.2 (15.2–25.5)	3.8 (2.8–4.8)	1.7 (1.2–2.2)	0.5 (0.4–0.7)
Overweight (25.0–29.9)	54 (15.0)	21.2 (17.5–26.1)	3.9 (3.1–4.9)	1.9 (1.5–3.0)	0.6 (0.4–0.7)
Obese (≥30.0)	25 (7.0)	17.4 (13.4–21.1)	3.5 (2.7–4.3)	1.3 (1.2–1.7)	0.5 (0.3–0.7)
Missing	35 (9.8)	17.3 (13.3–22.7)	3.5 (2.8–4.5)	1.6 (1.4–2.2)	0.5 (0.3–0.7)
Maternal education					
Low	61 (17.0)	18.4 (14.5–23.6)	3.6 (2.8–4.5)	1.7 (1.4–2.2)	0.6 (0.4–0.7)
Medium	115 (32.0)	19.6 (15.0–26.1)	3.7 (2.9–5.1)	1.6 (1.2–2.2)	0.6 (0.4–0.7)
High	170 (47.4)	20.4 (15.2–25.3)	4.0 (2.8–4.8)	1.7 (1.3–2.2)	0.5 (0.4–0.7)
Missing	13 (3.6)	15.8 (12.7–21.3)	3.3 (2.9–4.1)	1.5 (1.1–1.9)	0.4 (0.4–0.7)
Maternal race					
White	340 (94.7)	20.0 (15.3–25.5)	3.8 (2.9–4.8)	1.6 (1.3–2.2)	0.5 (0.4–0.7)
Nonwhite	7 (2.0)	14.6 (11.8–19.2)	2.4 (2.0–2.9)	1.4 (1.0–1.7)	0.5 (0.3–0.7)
Missing	12 (3.3)	17.1 (12.6–22.0)	3.5 (2.6–4.3)	1.7 (1.3–2.4)	0.4 (0.3–0.6)
Maternal age at delivery					
<25 years	77 (21.5)	18.7 (13.7–23.5)	3.9 (3.0–4.8)	1.6 (1.2–2.1)	0.5 (0.4–0.6)
25–29 years	141 (39.3)	20.9 (15.4–25.5)	3.8 (2.9–4.8)	1.6 (1.2–2.2)	0.6 (0.4–0.7)
≥30 years	141 (39.3)	19.5 (15.4–26.0)	3.6 (2.7–4.7)	1.7 (1.3–2.4)	0.6 (0.4–0.7)
Maternal smoking status					
Yes	72 (20.1)	17.2 (13.3–21.8)	3.5 (2.9–4.5)	1.7 (1.3–2.3)	0.5 (0.3–0.6)
No	274 (76.3)	21.1 (15.8–25.9)	3.8 (2.9–4.9)	1.6 (1.2–2.2)	0.6 (0.4–0.7)
Missing	13 (3.6)	16.0 (12.7–22.9)	3.6 (2.9–4.4)	1.6 (1.3–2.4)	0.6 (0.4–0.7)
Previous live birth					
Yes	176 (49.0)	18.1 (14.0–23.7)	3.1 (2.4–3.9)	1.5 (1.1–2.2)	0.5 (0.3–0.7)
No	170 (47.4)	22.1 (17.5–26.6)	4.5 (3.5–5.5)	1.8 (1.4–2.4)	0.6 (0.5–0.7)
Missing	13 (3.6)	16.0 (12.7–22.9)	3.6 (2.9–4.4)	1.6 (1.3–2.4)	0.6 (0.4–0.7)
Low birthweight (<2500 g at delivery)					
Yes	15 (4.2)	21.8 (17.5–38.2)	4.1 (3.3–5.6)	1.7 (1.4–2.3)	0.7 (0.4–0.7)
No	342 (95.8)	19.6 (14.9–25.1)	3.7 (2.8–4.8)	1.6 (1.2–2.2)	0.5 (0.4–0.7)
Missing	2 (0.56)	25.6 (22.7–28.5)	4.3 (4.2–4.4)	2.2 (1.6–2.7)	0.6 (0.6–0.7)
Preterm delivery (<37 weeks gestation)					
Yes	12 (3.3)	23.9 (14.8–33.6)	4.7 (3.1–5.5)	1.7 (1.4–2.0)	0.6 (0.4–0.7)
No	346 (96.4)	19.6 (15.0–25.2)	3.7 (2.9–4.8)	1.6 (1.3–2.2)	0.5 (0.4–0.7)
Missing	1 (0.3)	22.7 (22.7–22.7)	4.4 (4.4–4.4)	1.6 (1.6–1.6)	0.7 (0.7–0.7)

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Table 1. Characteristics of Study Population ($N = 359$) *continued*

	Frequency N (%)	PFOS (ng/mL)	PFOA (ng/mL)	PFHxS (ng/mL)	PFNA (ng/mL)
		Median (IQR)			
Ever breastfed					
Yes	278 (77.4)	19.7 (15.0–25.2)	3.7 (2.8–4.7)	1.6 (1.2–2.2)	0.5 (0.4–0.7)
No	61 (17.0)	21.7 (15.6–26.3)	4.2 (3.1–5.1)	1.7 (1.3–2.3)	0.5 (0.4–0.7)
Missing	20 (5.6)	18.6 (13.9–22.9)	3.4 (2.6–4.9)	1.7 (1.5–2.4)	0.6 (0.3–0.7)
Menarche (years)					
≥11.5	192 (53.5)	20.0 (14.7–25.4)	3.6 (2.7–4.7)	1.7 (1.2–2.3)	0.5 (0.4–0.7)
<11.5 (early)	167 (46.5)	19.6 (15.4–25.2)	3.9 (2.9–5.0)	1.6 (1.3–2.2)	0.6 (0.4–0.7)

IQR, interquartile range; PFHxS, perfluorohexane sulfonate; PFNA, perfluorononanoate; PFOA, perfluorooctanoate; PFOS, perfluorooctane sulfonate.

statistically significant, there were not meaningful associations within strata of education.

Results from our sensitivity analyses with BMI and WC (Table 2) generally supported the results observed for total %BF and %TF; for BMI and WC, we observed some significant inverse associations with PFOA and PFOS in main effects models. Consistent with the results for DXA body fatness measures, we also observed modification by mothers' educational status for PFOA and PFOS. Overall, the patterns of associations observed for PFASs with BMI and WC within strata of mother's educational status were similar to those observed for our DXA body fatness measures.

Discussion

Obesity is a multifactorial and complex disease and a number of dynamic epigenetic, environmental, lifestyle, and social interactions likely contribute to its etiology.[23] In this British cohort, we did not observe an overall association between maternal PFAS exposure and total %BF among young girls. Prenatal exposure to PFOA and PFOS was associated with total %BF in girls within some strata of maternal education status.

It is plausible that educational status is serving as a proxy for some other factor(s) that influence the associations between maternal PFAS concentrations and %BF among daughters. For example, educational attainment is a well-known marker for socioeconomic status, where disparities have been observed for maternal health-related behaviors and childhood obesity.[24,25]

Disparities may exist for exposures to PFASs. In the early 1990s when the ALSPAC prenatal blood collection took place, one important source of PFAS exposure is thought to have occurred through contact with new furnishings and carpets in home and office settings; thus, participants of higher socioeconomic status might have experienced the highest exposures. Interestingly, concentrations of maternal PFASs have been positively associated with educational status.[26–28]

In this study, maternal PFOS and PFOA tended to be higher among mothers reporting higher educational status (Table 1). Daughters of less educated (lower socioeconomic status) moms may experience more complex dynamic combinations of environmental, lifestyle, and social stressors over time.[23] Maternal education status is generally positively associated with diet quality during pregnancy and childhood but inversely associated with childhood obesity,[25,29] and in this study, daughters of mothers with less education tended to have higher body fatness measures (Supplementary Table S3). Families with more limited resources experience greater variability in some well-known contributors to childhood adiposity, including diet composition, diet quality, and acute and chronic food security, which are challenging to measure accurately (*i.e.*, self-reported) in epidemiologic research.[30] This could at least, in part, contribute to the inconsistencies we observed by maternal educational status.

Previous observational epidemiologic studies that have examined the role of PFASs in childhood weight have yielded divergent results overall and within studies results have lacked consistency across the PFASs measured, not unlike the results in this study. Andersen et al.[31] evaluated the role of prenatal exposure to PFOA and PFOS on measures of body fatness in 811 7-year-old children whose mothers participated in the Danish National Birth Cohort during 1996–2002. In contrast to the ALSPAC population, PFOA and PFOS plasma concentrations were not associated with lower weight in infancy in the Danish cohort.[32] Similarly, maternal plasma PFASs were not significantly associated with BMI and WC among 7-year-old girls. It is noteworthy that both PFOA (median 5.3 ng/mL) and PFOS (median 33.8 ng/mL) concentrations were higher in Danish prenatal samples than we observed in ALSPAC (Supplementary Table S1). Halldorsson et al.[33] assessed the association between prenatal PFAS serum concentrations and weight among 665 offspring ($N = 320$ males; 345 females) of mothers recruited into the Aarhus Denmark birth cohort (1988–1989). Median maternal PFAS concentrations in this

Table 2. Regression Coefficients (β) for the Relationship between Perfluoroalkyl Substances and Measures of Body Fatness Overall[a] and by Educational Status (Low, Medium, High)

	DXA-total body fat (%) ($N = 319$)			DXA-trunk fat (%) ($N = 319$)			BMI (kg/m^2) ($N = 312$)			WC (cm) ($N = 319$)		
Analyte	β	**95% CI**	***p***	β	**95% CI**	***p***	β	**95% CI**	***p***	β	**95% CI**	***p***
PFOS (ng/mL)												
Overall[b]	−0.07	−0.16 to 0.02	0.12	−0.06	−0.12 to −0.01	**0.02**	−0.04	−0.07 to 0.00	**0.03**	−0.12	−0.20 to −0.04	**0.005**
Low education	0.02	−0.17 to 0.22	0.81	−0.05	−0.17 to 0.06	0.37	−0.01	−0.08 to 0.06	0.76	−0.03	−0.21 to 0.16	0.78
Medium	0.09	−0.08 to 0.26	0.30	0.07	−0.03 to 0.18	0.17	0.02	−0.04 to 0.08	0.60	0.04	−0.12 to 0.20	0.62
High	−0.19	−0.31 to −0.07	**0.003**	−0.13	−0.21 to −0.06	**<0.001**	−0.07	−0.11 to −0.03	**0.001**	−0.23	−0.35 to 0.12	**<0.0001**
P-interactions			**0.02**			**0.006**			0.06			**0.01**
PFOA (ng/mL)												
Overall[b]	−0.30	−0.76 to 0.16	0.20	−0.27	−0.55 to 0.00	0.05	−0.16	−0.32 to 0.00	0.05	−0.54	−0.97 to −0.11	**0.01**
Low education	−1.03	−2.35 to 0.29	0.13	−0.51	−1.31 to 0.28	0.21	−0.63	−1.10 to −0.16	**0.009**	−1.29	−2.52 to −0.07	**0.04**
Medium	1.41	0.28 to 2.54	**0.01**	0.35	−0.33 to 1.04	0.31	0.41	0.02 to 0.81	**0.04**	1.16	0.11 to 2.21	**0.03**
High	−0.58	−1.12 to −0.04	**0.03**	−0.38	−0.70 to −0.05	**0.02**	−0.22	−0.41 to −0.03	**0.02**	−0.82	−1.32 to −0.32	**0.001**
P-interactions			**0.005**			0.14			**0.003**			**0.002**
PFHxS (ng/mL)[c]												
Overall[b]	−0.06	−0.21 to 0.09	0.47	−0.01	−0.11 to 0.08	0.77	−0.02	−0.08 to 0.03	0.37	−0.08	−0.22 to 0.06	0.28
Low education	−3.09	−6.04 to −0.13	**0.04**	−1.48	−3.25 to 0.29	0.10	−1.22	−2.29 to −0.15	**0.03**	−1.45	−4.23 to 1.34	0.31
Medium	0.17	−0.21 to 0.54	0.38	0.19	−0.03 to 0.42	0.09	0.02	−0.11 to 0.15	0.80	−0.03	−0.38 to 0.33	0.88
High	−0.09	−0.26 to 0.07	0.28	−0.05	−0.15 to 0.05	0.32	−0.03	−0.09 to 0.03	0.33	−0.09	−0.24 to 0.07	0.28
P-interactions			0.06			**0.04**			0.08			0.60
PFNA (ng/mL)												
Overall[b]	1.71	−1.29 to 4.71	0.26	−0.03	−1.83 to 1.77	0.97	0.22	−0.83 to 1.27	0.68	−0.11	−2.91 to 2.70	0.94
Low education	12.73	0.11 to 25.36	**0.04**	−0.97	−8.58 to 6.63	0.80	4.06	−0.53 to 8.65	0.08	3.26	−8.62 to 15.14	0.59
Medium	3.43	−1.95 to 8.81	0.21	2.13	−1.11 to 5.37	0.20	0.71	−1.17 to 2.60	0.46	1.80	−3.26 to 6.86	0.49
High	0.12	−3.53 to 3.77	0.95	−0.94	−3.14 to 1.26	0.40	−0.26	−1.54 to 1.02	0.69	−1.22	−4.66 to 2.22	0.49
P-interactions			0.13			0.30			0.17			0.53

[a]Models adjusted for sampling design, prepregnancy BMI (kg/m^2) and maternal educational status. PFNA models were also adjusted for maternal smoking status.

[b]Main effects model without the interaction terms.

[c]There was one fewer sample analyzed for PFHxS; all n's for PFHxS are one less than for PFOA and PFOS. PFNA models $N = 313$ due to missing smoking data except for the BMI model where $N = 306$.

95% CI, 95% confidence interval; DXA, dual-energy X-ray absorptiometry; WC, waist circumference.

study were similar to ours (Table 1): PFOA was 3.7 ng/mL, PFOS was 21.5 ng/mL, and PFNA was 0.3 ng/mL. In the Aarhus study, maternal PFOA concentrations were positively and significantly associated with BMI and WC among females at 20 years of age. Similar, but weaker, associations were observed for PFOS and PFNA among girls. Hoeyer et al.[34] analyzed PFOA and PFOS from the sera of 1022 pregnant women from Greenland and the Ukraine, enrolled in the INUENODO cohort between 2002 and 2004. Median concentrations of maternal PFOA and PFOS were higher among the Greenland sample (median PFOA 1.8 ng/mL; median PFOS 20.2 ng/mL) and more similar to our population, than those from the Ukraine sample (median PFOA 1.0 ng/mL; median PFOS 5.0 ng/mL).

The investigators reported adjusted relative risk [RR (95% CI)] of offspring overweight (defined as >85 percentile for age and sex) and unfavorable offspring waist-to-height ratio (>0.5) for continuous (natural log transformed) and tertiles of PFASs between ages 5–9 years. In country-specific and in pooled analyses, neither PFOA nor PFOS was significantly associated with offspring risk of overweight.[31] In the pooled analysis, continuous PFOS concentration was positively associated with offspring waist-to-height ratio (RR 1.38; 95% CI: 1.05 to 1.82); however, in gender-stratified analyses, results were statistically significant only for girls (RR 1.54; 95% CI: 1.06 to 2.23). There were no statistically significant findings for the association of tertiles of maternal PFOA with waist-to-height ratios.[34]

In the Cincinnati-based HOME Study, Braun et al.[35] evaluated associations of prenatal PFAS concentrations (PFOA, PFOS, PFHxS, and PFNA) with BMI, WC, and %BF (measured by a Tanita body fat monitor) among a modestly sized sample of 8-year-old children of both sexes ($N = 204$). Median prenatal PFOA concentration in HOME was 5.3 ng/mL, nearly 1.5 times as high as in our population (median 3.7 ng/mL). This study reported positive associations between PFOA and BMI z-score trajectories, WC, and %BF. Results were not reported by sex; however, the results stated that sex did not modify the associations. No associations were observed for the other PFASs measured (PFOS, PFHxS, and PFNA). The investigators noted that their study was limited by the use of less-refined measures of body fatness and that future studies should take advantage of more comprehensive measures such as DXA.[35] In this study, we observed positive associations between PFOA and BMI, WC and DXA-derived total %BF, but only among daughters of mothers in the middle education group.

Finally, in the only other study to date that has evaluated the association between prenatal PFAS exposure and body fatness measured by DXA in school-age girls, Mora et al.,[36] affiliated with Project Viva, observed small and relatively consistent nonsignificant positive associations of prenatal PFASs (PFOS, PFOA, PFHxS, and PFNA) with DXA-derived total and trunk fat mass indices (kg/m^2) among 344 girls.[36] Results for %BF and %TF were not reported.

In a recent review, Vandenberg et al.[37] discuss mechanisms for a variety of observed responses to low-dose exposure to EDCs. For example, even at low doses, EDCs that influence the production, release, transport, binding, or metabolism (*e.g.*, degradation) of natural hormones altering their availability could lead to biologically significant effects.[37]

Prenatal PFAS exposure might influence childhood body fatness among girls through a number of potential mechanisms.[3,5,38] According to a recent Scientific Statement on EDCs issued by the Endocrine Society,[39] EDCs have broad effects on numerous endocrine endpoints *in vivo* and may act through an array of mechanisms, including actions on estrogen and androgen receptors and through thyroid effects.

In animal models, a growing body of research suggests that exposure to EDCs can increase the number and size of adipocytes, alter insulin metabolism, and disrupt energy balance.[40] While the molecular mechanisms involved are unclear, some research suggests that EDCs can interfere with epigenetic programming of gene regulation through activation of fat-regulating nuclear receptors such as peroxisome proliferator-activated receptor gamma, which can lead to weight gain.[41–43] Other potential mechanisms could include effects on neurocognitive development that could alter the processing of sensory information related to ingestive behaviors throughout life.[44]

There are a number of strengths to our analyses. All the data for this study were collected prospectively by trained staff under tightly controlled conditions. PFASs were measured at the National Center for Environmental Health laboratories of the CDC, the laboratory that measures PFASs for NHANES. Concentrations of PFASs in the prenatal samples in our study are similar to those reported in recent analysis of samples from the 2009 to 2010 NHANES; thus, our results are potentially of relevance to current US populations.[45] Body fatness (composition) was measured by DXA, which provides a more accurate estimate of fat mass than BMI or WC.[46]

There are also some potential limitations. Maternal PFAS concentrations were measured only once during pregnancy over a range of gestational ages, and daughters' PFAS concentrations measured postnatally are not available. Generalizability of our findings was reduced by the demographic characteristics (mainly Caucasian) and geography (only UK) of the cohort.

Our analyses were conducted on a sample of mother–daughter dyads selected for an ancillary study of pubertal development (boys were not included in this analysis); weighted linear regression models were used to adjust for the sampling scheme (results stratified by age of menarche included as Supplementary Table S4). This study includes a subset of the daughters included in the ancillary puberty development study who completed DXA scans for the assessment of body fatness (359 of 448); thus, selection bias is a potential consideration. Compared to the overall ALSPAC cohort, those children who had DXA scans at age 9 tended to have mothers who were more educated and less likely to have smoked during pregnancy; however, there were no significant differences in BMI by DXA participation.[21] If the girls included in our analyses were not representative of

all ancillary study participants or of the parent study, then our results could be biased. As reported previously, maternal characteristics for girls included in the ancillary sample ($N=448$) were similar to the group of girls enrolled in the cohort.[13] For example, ~20% of mothers were in the lowest educational group in the overall cohort and in the ancillary sample; 25% of all mothers reported prenatal smoking compared to 23% in the ancillary sample.

Our sample with complete data for both prenatal PFASs and DXA scans at age 9 years ($N=359$) was relatively representative of the overall ancillary sample,[12] although it was likely of somewhat higher socioeconomic status. For example, only 17% of mothers in this study were in the lowest education group and 20% reported smoking during pregnancy. This study comprised 46.5% of girls with early menarche compared to 48.8% for the full ancillary study. We evaluated numerous potential confounders, but as in most research, we cannot rule out the possibility of residual confounding. Finally, similar to other previous investigations, our results are modest and do not show a consistent pattern of direction or magnitude of association across the PFASs measured.

Conclusions

In conclusion, in this British cohort, we did not observe an overall association between maternal PFAS exposure and total %BF among young girls. Prenatal exposure to PFOA and PFOS was associated with total %BF in girls within some strata of maternal education status. The results for associations of PFOA and PFOS with other measures of body fatness were generally reflective of those observed for total %BF. We did not observe strong associations for PFHxS or PFNA with measures of body fatness. The role of environmental chemicals, such as PFASs, in the development of overweight and obesity at different life stages is an important topic of emerging investigation, and additional research is needed in this area.

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Author Disclosure Statement

No competing financial interests exist.

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