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# Association of perfluoroalkyl substances with gestational hypertension and preeclampsia in the MIREC study

Michael M. Borghese[a,*], Mark Walker[b], Michael E. Helewa[c], William D. Fraser[d], Tye E. Arbuckle[a]

[a] Environmental Health Science and Research Bureau, Health Canada, Ottawa, ON K1A 0K9, Canada
[b] Department of Obstetrics, Gynecology and Newborn Care, The Ottawa Hospital, Ottawa, ON, Canada
[c] Department of Obstetrics, Gynecology and Reproductive Sciences, University of Manitoba, Winnipeg, MB, Canada
[d] Department of Obstetrics and Gynecology, University of Sherbrooke, Sherbrooke, QC, Canada

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ABSTRACT

*Background:* Perfluoroalkyl substances (PFAS) have been linked with a number of developmental, reproductive, hepatic, and cardiovascular health outcomes. However, the evidence for an association between PFAS and hypertensive disorders of pregnancy (including gestational hypertension and preeclampsia) is equivocal and warrants further investigation.

*Objectives:* To examine the relationship between background levels of perfluorooctanoic acid (PFOA), perfluorooctane sulfonate (PFOS), and perfluorohexane sulfonate (PFHxS) and the development of gestational hypertension or preeclampsia in a Canadian pregnancy cohort. We also explored the potential for effect modification according to fetal sex.

*Methods:* Maternal plasma samples were collected in the first trimester from participants in the MIREC study and were analyzed for PFOA, PFOS, and PFHxS. Blood pressure was measured during each trimester. Gestational hypertension and preeclampsia were defined using the Society of Obstetricians and Gynaecologists of Canada guidelines. Logistic regression models were used to derive adjusted odds ratios (OR) and 95% confidence intervals (CI) for associations between PFAS concentrations (per doubling of concentration as well as according to tertiles) and gestational hypertension or preeclampsia. Linear mixed models were used to examine the association between PFAS concentrations and changes in blood pressure throughout pregnancy.

*Results:* Data from 1739 participants were analyzed. 90% of women were normotensive throughout pregnancy, 7% developed gestational hypertension without preeclampsia, and 3% developed preeclampsia. In the full analyses, neither PFOA nor PFOS were associated with gestational hypertension or preeclampsia. However, each doubling of PFHxS plasma concentration was associated with higher odds of developing preeclampsia (OR = 1.32; 95% CI: 1.03, 1.70). In addition, participants in the highest PFHxS tertile (1.4–40.0 μg/L) had higher odds of developing preeclampsia relative to those in the lowest tertile (OR = 3.06; 95% CI: 1.27, 7.39). In stratified analyses, this effect was only apparent among women carrying a female fetus (OR = 4.90; 95% CI: 1.02, 22.3). However, among women carrying a male fetus, both PFOS and PFHxS were associated with gestational hypertension, but not preeclampsia. Higher plasma concentrations of all three PFAS were associated with increases in diastolic blood pressure throughout pregnancy, and PFOA and PFHxS were also associated with systolic blood pressure. Discrepant findings were similarly revealed in analyses stratified by fetal sex.

*Conclusions:* Higher levels of PFHxS were associated with the development of preeclampsia, but not gestational hypertension. Neither PFOA nor PFOS were associated with either outcome. However, we show, for the first time, that fetal sex may modify these associations, a finding which warrants replication and further study.

## 1. Introduction

Perfluoroalkyl substances (PFAS), such as perfluorooctanoic acid (PFOA), perfluorooctane sulfonate (PFOS), and perfluorohexane sulfonate (PFHxS), are a class of structurally related chemicals containing a highly fluorinated carbon backbone that have been produced since the 1950s (Buck et al., 2011). The strong C—F bonds provide these substances with considerable thermal and chemical stability and their amphipathic nature allows them to effectively repel water and oils. These properties make PFAS useful for manufacturing a wide range of

* Corresponding author.
*E-mail address:* Michael.borghese@canada.ca (M.M. Borghese).

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consumer products (Health Canada, 2013), including non-stick cookware, food packaging, personal care and beauty products, fire retardant foams, and carpet treatment applications, among others. Food, drinking water (Trudel et al., 2008), and household dust (i.e., from carpet treatment applications) (Beesoon et al., 2012), are thought to be the primary sources of exposure. Because of their remarkable stability, these substances are persistent in the environment and have been found worldwide in human (Glynn et al., 2012; Haines et al., 2017; Health Canada, 2013; Kannan et al., 2004; Kärrman et al., 2006; Kato et al., 2011) and wildlife (Houde et al., 2006) populations. In fact, human exposure to these substances is ubiquitous (98–100% of the Canadian population have detectable levels in their blood (Haines et al., 2017)) and since they are well-absorbed, minimally metabolized, and poorly excreted (Harada et al., 2005; Kennedy et al., 2004), they tend to bioaccumulate; the geometric mean half-lives range from 3.5 (PFOA) to 7.3 (PFHxS) years (Olsen et al., 2007). As a result, some of these substances have received considerable research and regulatory attention, most notably PFOA, PFOS, and PFHxS. Since 2002, the voluntary phase-out of manufacturing of PFOS and related substances in the US is thought to have contributed to observed declines in population levels of PFOS and PFOA (Calafat et al., 2007; Glynn et al., 2012; Kato et al., 2011); however, temporal trends for other PFAS (including PFHxS) are less consistent and in some cases increases have been observed (Glynn et al., 2012; Kato et al., 2011).

PFAS (mainly PFOA and PFOS) have been linked with a number of developmental, reproductive, hepatic, and cardiovascular health outcomes (Andersen et al., 2008; Kennedy et al., 2004; Lau et al., 2007). One of these potential effects is on hypertensive disorders of pregnancy, including gestational hypertension (elevated systolic and/or diastolic blood pressure) and preeclampsia (elevated blood pressure accompanied by related complications, such as proteinuria) (Magee et al., 2014). These conditions are associated with higher risk of maternal, fetal, and newborn morbidity and mortality (Butalia et al., 2018; Jelin et al., 2009; Lo et al., 2013; Nakimuli et al., 2016; van Esch et al., 2017; Ying et al., 2018). In Canada, the prevalence of gestational hypertension and preeclampsia in 2010/2011 was 46.2 and 11.5 per 1000 deliveries, respectively (Public Health Agency of Canada, 2014). This rate has remained relatively stable in Canada since 2004/2005, but data from the US suggest that the prevalence of these conditions is considerably higher than in the preceding two decades (Wallis et al., 2008). While there has been a shift towards more inclusive definitions of preeclampsia, encompassing associated complications and thus increasing the number of diagnoses, this likely does not fully explain this increase in prevalence over time.

The pathophysiology underlying the development of preeclampsia, as well as the role of environmental chemicals such as PFAS, is not entirely understood. Preeclampsia is generally believed to be a two-stage disease whereby defects in spiral artery remodelling and placental cytotrophoblast invasion play a key role in early disease progression (i.e., the first, or placental, stage) (Pijnenborg et al., 2006; Steegers et al., 2010). This defective remodelling results in placental ischemia via hypoperfusion, which leads to abnormal placental stress responses (i.e., generation of reactive oxygen species) and ultimately to the apoptotic release of pro-inflammatory trophoblast and syncytial debris into the maternal circulation (Huppertz, 2008; Huppertz et al., 2003). This cascade of events can, but does not always, lead to a systemic maternal inflammatory response that is characteristic of the second (i.e., maternal) stage of preeclampsia. PFAS have been shown to be toxic to human placental cytotrophoblast cells by interfering with lipid homeostasis and metabolism, particularly for cell membrane lipids, and have also been shown to inhibit aromatase activity in these same cells (Gorrochategui et al., 2014). Thus, PFAS could contribute to defective spiral artery remodelling by interfering with cytotrophoblast invasion, and this has the potential to be fetal sex-specific. Previous studies have demonstrated inconsistent fetal sex-specific effects of PFAS on some perinatal outcomes (Andersen et al., 2010; Wikström et al., 2019a).

The epidemiological evidence for associations between PFAS and hypertensive disorders of pregnancy is equivocal, with eight studies demonstrating a mixture of null and weak positive findings, often with inconsistent dose–response patterns. Five of these studies examined a population of pregnant women from the Mid-Ohio Valley region of the United States that was exposed to high levels of PFOA (Darrow et al., 2013; Nolan et al., 2010; Savitz et al., 2012a, 2012b; Stein et al., 2009). In some of these studies, null findings could have been impacted by misclassification of exposures that were derived using ZIP codes in relation to public water service facilities (Nolan et al., 2010) or were retrospectively reconstructed using residential information (Savitz et al., 2012a, 2012b). In contrast, positive associations with preeclampsia were observed when serum concentrations of PFOA and PFOS were measured (Darrow et al., 2013; Stein et al., 2009). However, these studies were limited by the potential for recall bias because self-reported pregnancy outcomes occurred prior to blood sampling and participants were aware of their exposure status. Moreover, all of the above studies relied on self-reported or birth certificate measures of hypertensive disorders of pregnancy, which are prone to error (DiGiuseppe et al., 2002; Stuart et al., 2013). In addition, these studies often did not distinguish preeclampsia from gestational hypertension, did not examine associations with measured blood pressure, and other PFAS (i.e., PFHxS) were not considered. Finally, evidence from the highly exposed Mid-Ohio Valley population may not be generalizable to other populations. Three studies of general obstetric populations have demonstrated mostly null results for PFOA, PFOS, and PFHxS (Huang et al., 2019; Starling et al., 2014; Wikström et al., 2019b), with some conflicting results for other PFAS. However, these studies were limited in that the authors examined preeclampsia only (not gestational hypertension or blood pressure) (Starling et al., 2014; Wikström et al., 2019b), excluded multiparous women (Starling et al., 2014), or used cord blood concentrations of PFAS which is limited in establishing temporality (Huang et al., 2019).

Given the equivocal evidence to date for PFOA and PFOS, and the limited evidence for PFHxS, there is a need to further examine potential associations between exposure to background levels of these PFAS and hypertensive disorders of pregnancy. Accordingly, the objective of this paper was to examine the relationship of PFOA, PFOS, and PFHxS with the development of gestational hypertension and preeclampsia, as well as serial measures of blood pressure, in a cohort of pregnant Canadian women. In addition, since carrying a male fetus may be associated with a higher likelihood of developing preeclampsia (Vatten and Skjærven, 2004), we also explored the potential for effect modification by fetal sex.

## 2. Methods

### 2.1. Study design and participants

This paper uses data from the Maternal-Infant Research on Environmental Chemicals (MIREC) study, which is a longitudinal Canadian pregnancy cohort study. The MIREC study was established to examine the potential adverse health effects of prenatal exposure to priority environmental chemicals on pregnancy and infant/child health. Potential health effects of PFAS, including associations with fecundity (Velez et al., 2015) and gestational weight gain (Ashley-Martin et al., 2016), have previously been studied in this cohort of pregnant women. A complete description of the MIREC study population is available elsewhere (Arbuckle et al., 2013). Briefly, 2001 pregnant women were recruited from 10 study sites across Canada during the first trimester between 2008 and 2011. Women were excluded if they had a serious medical condition, were < 18 years old, > 14 weeks gestation, or could not communicate in either English or French. Participants completed baseline and subsequent questionnaires to gather socio-demographic and exposure information. Clinical data were abstracted from medical charts. Women who withdrew from the study

(n = 18) or experienced a miscarriage or stillbirth (n = 74) were excluded from this analysis, leaving an eligible sample size of 1 909 participants. In the present analysis, participants with missing data for PFAS (n = 39) or blood pressure (n = 32), as well as those with chronic hypertension (n = 57), or carrying multiple fetuses (n = 42) were excluded which resulted in a final analytical sample of 1 739 participants. This study was approved by Health Canada's Research Ethics Board, as well as the Research Ethics Committees of Sainte-Justine University Hospital in Montreal, Canada and those of all 10 MIREC-affiliated study sites. Informed consent was obtained from all participants.

### 2.2. Measurement of PFAS in plasma

Maternal blood samples were collected during a first-trimester clinic visit using 10-ml sterile vacutainer tubes. The mean (range) of gestational age in weeks at the blood draw was 11.6 (2–14). Within 2 h of the blood draw, samples were centrifuged and the plasma was aliquoted into smaller cryovials and stored at −80 °C. Analysis of PFAS was performed by the Laboratoire de Toxicologie, Institut National de Santé Publique du Québec (Quebec City, Quebec, Canada), which is accredited by the Standards Council of Canada. This was done using ultra-high-pressure liquid chromatography (ACQUITY UPLC System; Waters Corporation, Milford, Massachusetts) coupled with tandem mass spectrometry, operated in the multiple reaction monitoring mode, with an electrospray ion source in negative mode. The between-assay coefficients of variation for PFOA, PFOS, and PFHxS were 5.8, 3.6, and 10.0%, respectively. The limits of detection (LODs) for PFOA, PFOS, and PFHxS were 0.1, 0.3, and 0.2 μg/L, respectively. Measures of these PFAS in maternal plasma during the 1st and 3rd trimesters in a subsample of women from the MIREC study were highly correlated (ICC range = 0.64–0.83) (Fisher et al., 2016), but 3rd trimester values were lower than 1st trimester values, which is consistent with previous research (Fei et al., 2007; Glynn et al., 2012; Papadopoulou et al., 2015).

### 2.3. Blood pressure measurement and definitions of gestational hypertension and preeclampsia

Systolic and diastolic blood pressure (SBP and DBP, respectively) were assessed by clinical staff using a sphygmomanometer at three prenatal clinic visits (at 6–13, 16–21, and 32–34 weeks). During the measurement, participants maintained a sitting position with the cuffed arm resting at the level of the heart. Two measures were recorded approximately one minute apart and were averaged. The Korotkoff phase V was used for DBP measurement. The Korotkoff phase IV was used only when a phase V was absent. SBP and DBP were also measured upon admission for delivery as well as 4 h post-delivery, and these values were abstracted from medical records. Clinical staff were blinded to the results of the chemical analysis for PFAS.

Using the Society of Obstetricians and Gynaecologists of Canada guidelines (Magee et al., 2014), a woman was considered as having gestational hypertension if SBP was ≥140 mmHg and/or DBP was ≥90 mmHg at a gestational age of 20 weeks or later, including upon admission for, but prior to, delivery. Women with gestational hypertension may or may not have received antihypertensive agents to control the hypertension. A participant was considered as having pre-existing (i.e., chronic) hypertension if hypertension was present at a gestational age of less than 20 weeks. Preeclampsia was defined as gestational hypertension with the addition of either: (1) proteinuria (defined as protein dipstick test ≥1 + OR proteinuria in 24-urine ≥300 mg/24 h or ≥0.3 g/L) or (2) related maternal complications (including disseminated intravascular coagulation, pulmonary edema, convulsions–eclampsia, transfusion, elevated liver enzyme levels, and/or platelet count < 50 × $10^9$/L). Participants who did not meet any of these conditions were considered to be normotensive. Gestational age in weeks was based on last menstrual period and/or early ultrasound result. Participants were categorized into three outcome groups for the current analysis: normotensive, gestational hypertension without preeclampsia, and preeclampsia. Previous studies examining potential associations with PFAS have defined preeclampsia as the presence of gestational hypertension and proteinuria only (Huang et al., 2019; Starling et al., 2014; Wikström et al., 2019b), consistent with clinical guidelines from the American College of Obstetricians and Gynaecologists (American College of Obstetricians and Gynecologists, 2002). In light of this, we conducted a sensitivity analysis using this restricted definition of preeclampsia to better contextualize our results within the literature.

### 2.4. Covariates

Several covariates were considered as *a priori* potential confounding variables in multivariable models. These were based on previous assessments of risk factors for gestational hypertension and preeclampsia among Canadian women (Shen et al., 2017), suspected predictors of plasma concentrations of PFAS among pregnant women (Berg et al., 2014; Brantsæter et al., 2013; Fisher et al., 2016; Lewin et al., 2017) as well as previous reports of possible associations between PFAS and hypertensive disorders of pregnancy (Darrow et al., 2013; Nolan et al., 2010; Savitz et al., 2012a, 2012b; Starling et al., 2014; Stein et al., 2009). Covariates included: maternal age at delivery in years (continuous), parity (nulliparous vs. multiparous), education (university, college, less than college), maternal smoking (never, quit before pregnancy, quit during pregnancy, currently smoking), and pre-pregnancy body mass index (BMI) (under/normal weight, overweight, obese). Few participants were underweight (n = 47), so participants with underweight and normal weight were grouped together in all analyses.

### 2.5. Statistical analysis

All analyses were performed using SAS Enterprise Guide 7.1 (SAS institute, Cary, NC). Descriptive statistics for participant characteristics are presented using frequencies or arithmetic means (SD), as appropriate. Plasma concentrations of PFAS were skewed and are presented using medians and inter-quartile ranges according to study outcome group and compared using Kruskal-Wallis ANOVA. Geometric means (95% CI) are provided for comparison with the literature. Prior to analysis, PFAS were transformed by log base 2 ($Log_2$) to normalize the distribution and to allow for interpretation of parameter estimates as per a doubling of concentration. Values below the limit of detection for PFOA (n = 3), PFOS (n = 3), and PFHxS (n = 68) were assigned a value of LOD/2. Spearman correlations were used to examine the relationships between PFAS.

Logistic regression models were used to calculate adjusted odds ratios (OR) and 95% confidence intervals (95% CI) for the association between PFAS and the development of either gestational hypertension or preeclampsia, relative to having a normal blood pressure. Analyses were conducted with PFAS as a $log_2$-transformed variable as well as grouped into tertiles, adjusting for aforementioned covariates. Missing covariate data were replaced using multiple imputation (Rubin, 1987) (PROC MI, m = 5) (White et al., 2011). Parameter estimates obtained from each of the iterations were combined using PROC MIANALYZE to provide appropriate variance estimation.

Linear mixed models were used to estimate the effect of $log_2$-transformed PFAS on changes in SBP and DBP throughout pregnancy. An AR(1) covariance matrix was used to account for the random effect of clustering of repeated observations within individuals. In addition, general linear models were used to estimate the association between $log_2$-transformed PFAS and SBP and DBP at each trimester. All models were adjusted for the aforementioned covariates. Linearity and dose–response patterns were assessed by inspecting predicted values and by using restricted cubic splines with knots at the 5th, 25th, 50th, 75th, and 95th percentiles (Desquilbet and Mariotti, 2010). Models generally

did not deviate from linearity and therefore results from the more parsimonious linear models are presented; however, in one model a non-linear dose- response pattern was observed and these results are presented for this model only. Regression diagnostics were used to assess other model assumptions. Since the use of antihypertensive medications would directly impact participants' SBP/DBP measures and could mask potential associations with PFAS, participants who reported using these medications during pregnancy (n = 31) were excluded from the analyses for SBP and DBP. These participants were not excluded the categorical analyses for gestational hypertension and preeclampsia.

We examined the potential for effect modification by fetal sex through stratified analyses as well as by investigating associations for $log_2$ PFAS-fetal sex interaction terms. Ultimately, the number of women who developed preeclampsia in this sample was low, and in this context these analyses should be considered exploratory. Although parity is another important factor to consider for effect modification, this was not possible in our sample because only 14 women who were multiparous developed preeclampsia.

## 3. Results

Participant characteristics (n = 1739) are presented in Table S1 according to study outcome group. Women were predominately Caucasian (85% of women) and their mean (SD) age was 32.1 (5.1) years. 1563 (89.9%) women were normotensive, 127 (7.3%) developed gestational hypertension (without preeclampsia), and 49 (2.8%) developed preeclampsia. Of the 49 women who developed preeclampsia, 36 had proteinuria and 13 did not. Other complications experienced by these 13 women included elevated liver enzymes (n = 9), blood transfusion (n = 2), pulmonary embolism (n = 1), and severe hypertension (SBP ≥ 160 mmHg and/or DBP ≥ 110 mmHg) (n = 1).

Table S2a contains the first trimester plasma concentrations (μg/L) of PFAS as well as SBP and DBP during each trimester as well as upon admission for, but prior to, delivery according to study outcome group. The largest difference between study outcome groups was observed for PFHxS: median PFHxS concentrations were 50% higher among those with preeclampsia vs. normal blood pressure. The three PFAS were moderately correlated with one another (all p < 0.0001): PFOA and PFOS (r = 0.56), PFOA and PFHxS (r = 0.49), PFOS and PFHxS (r = 0.54). Geometric means (95% CI) in μg/L for PFOA, PFOS, and PFHxS were 1.65 (1.61, 1.70), 4.56 (4.44, 4.69), and 1.02 (0.98, 1.06), respectively. SBP and DBP were consistently higher among those with gestational hypertension or preeclampsia vs. normal blood pressure. While the SBP and DBP of those with normal blood pressure were similar across trimesters, values among those with gestational hypertension or preeclampsia were similar between trimesters 1 and 2, increased during trimester 3, and further increased upon admission for delivery. Plasma concentrations (μg/L) of PFAS according to study outcome group, and stratified by fetal sex, are provided in Tables S2b and S2c. The direction and magnitude of differences were consistent with those from the full sample with one exception: among women carrying male infants, those with gestational hypertension had higher values of PFOS than those with normal blood pressure or preeclampsia. Table S4 contains the highest recorded values for SBP and DBP overall and stratified by fetal sex, according to outcome group.

In logistic regression models, first trimester plasma concentrations of PFOA and PFOS were not associated with the odds of developing gestational hypertension or preeclampsia when examined both linearly and by tertile (Table 1a). Similarly, results suggest that PFHxS was not associated with the odds of developing gestational hypertension. However each doubling of PFHxS plasma concentration was associated with higher odds of developing preeclampsia (OR = 1.32; 95% CI: 1.03, 1.70). Participants in the highest PFHxS tertile (1.4–40.0 μg/L) had higher odds of developing preeclampsia relative to those in the lowest tertile (OR = 3.06; 95% CI: 1.27, 7.39), although the association was observed for those in the middle tertile was not statistically significant. When we applied a more restrictive definition of preeclampsia (gestational hypertension and proteinuria only), effect estimates for all three PFAS were attenuated and none were found to be associated with the development of preeclampsia (Table S3).

The results for analyses of gestational hypertension were discrepant when stratified by fetal sex (Tables 1b and 1c). Among women carrying male fetuses, each doubling of concentration of PFOS (OR = 1.46; 95% CI: 1.05, 2.01) or PFHxS (OR = 1.31; 95% CI: 1.05, 1.63) was associated with higher odds of developing gestational hypertension in linear models; however, results were not statistically significant when

**Table 1a**
Adjusted OR (95% CI) for the association between PFOA, PFOS, or PFHxS and gestational hypertension (without preeclampsia) or preeclampsia among pregnant women (n = 1739).

| | Gestational hypertension | | Preeclampsia | |
|---|---|---|---|---|
| | OR (95% CI) | p-value | OR (95% CI) | p-value |
| *PFOA* | | | | |
| $Log_2$ continuous | 1.06 (0.84, 1.35) | 0.61 | 1.36 (0.90, 2.08) | 0.15 |
| Tertile 1 (0.05–1.3) | ref | – | Ref | – |
| Tertile 2 (1.4–2.1) | 0.89 (0.55, 1.43) | 0.63 | 1.62 (0.69, 3.74) | 0.27 |
| Tertile 3 (2.2–11.0) | 1.13 (0.68, 1.86) | 0.64 | 1.23 (0.50, 3.00) | 0.65 |
| *PFOS* | | | | |
| $Log_2$ continuous | 1.15 (0.91, 1.45) | 0.24 | 1.25 (0.84, 1.82) | 0.28 |
| Tertile 1 (0.15–3.7) | ref | – | ref | – |
| Tertile 2 (3.8–5.8) | 1.43 (0.90, 2.29) | 0.13 | 1.72 (0.77, 3.82) | 0.19 |
| Tertile 3 (5.9–36.0) | 1.38 (0.84, 2.23) | 0.19 | 1.55 (0.68, 3.49) | 0.29 |
| *PFHxS* | | | | |
| $Log_2$ continuous | 1.15 (0.98, 1.35) | 0.09 | 1.32 (1.03, 1.70) | 0.03 |
| Tertile 1 (0.10–0.77) | ref | – | ref | – |
| Tertile 2 (0.78–1.3) | 1.03 (0.64, 1.67) | 0.91 | 1.40 (0.54, 3.63) | 0.48 |
| Tertile 3 (1.4–40.0) | 1.39 (0.87, 2.20) | 0.17 | 3.06 (1.27, 7.39) | 0.01 |

Plasma concentrations for tertiles are in μg/L.
OR – odds ratios; 95% CI – 95% confidence intervals.
Odds ratios for continuous variables represent a doubling (per $log_2$ increase) in plasma concentration.
Models are adjusted for maternal age, education, smoking status, pre-pregnancy BMI, and parity.

**Table 1b**
Adjusted OR (95% CI) for the association between PFOA, PFOS, or PFHxS and gestational hypertension (without preeclampsia) or preeclampsia among pregnant women carrying male fetuses (n = 913).

| | Gestational hypertension | | Preeclampsia | |
|---|---|---|---|---|
| | OR (95% CI) | p-value | OR (95% CI) | p-value |
| *PFOA* | | | | |
| $Log_2$ continuous | 1.26 (0.90, 1.75) | 0.18 | 1.23 (0.70, 2.16) | 0.46 |
| Tertile 1 (0.05–1.3) | ref | – | ref | – |
| Tertile 2 (1.4–2.1) | 1.12 (0.58, 2.16) | 0.74 | 0.86 (0.28, 2.66) | 0.80 |
| Tertile 3 (2.2–11.0) | 1.55 (0.78, 3.00) | 0.21 | 1.00 (0.32, 3.10) | 0.99 |
| *PFOS* | | | | |
| $Log_2$ continuous | 1.46 (1.05, 2.01) | 0.02 | 1.19 (0.72, 1.97) | 0.50 |
| Tertile 1 (0.15–3.7) | ref | – | ref | – |
| Tertile 2 (3.8–5.8) | 1.43 (0.75, 2.77) | 0.27 | 0.90 (0.31, 2.59) | 0.84 |
| Tertile 3 (5.9–36.0) | 1.80 (0.94, 3.46) | 0.07 | 1.08 (0.38, 3.10) | 0.88 |
| *PFHxS* | | | | |
| $Log_2$ continuous | 1.31 (1.05, 1.63) | 0.02 | 1.32 (0.92, 1.88) | 0.12 |
| Tertile 1 (0.10–0.77) | ref | – | ref | – |
| Tertile 2 (0.78–1.3) | 1.21 (0.62, 2.34) | 0.58 | 1.15 (0.34, 3.94) | 0.83 |
| Tertile 3 (1.4–40.0) | 1.72 (0.92, 3.19) | 0.09 | 2.41 (0.81, 7.17) | 0.12 |

OR – odds ratios; 95% CI – 95% confidence intervals.
Odds ratios for continuous variables represent a per $log_2$ increase in perfluoroalkyl substance plasma concentration.
Models are adjusted for maternal age, education, smoking status, pre-pregnancy BMI, and parity.

**Table 1c**
Adjusted OR (95% CI) for the association between PFOA, PFOS, or PFHxS and gestational hypertension (without preeclampsia) or preeclampsia among pregnant women carrying female fetuses (n = 824).

| | Gestational hypertension | | Preeclampsia | |
|---|---|---|---|---|
| | OR (95% CI) | p-value | OR (95% CI) | p-value |
| *PFOA* | | | | |
| $Log_2$ continuous | 0.87 (0.61, 1.23) | 0.43 | 1.54 (0.81, 2.94) | 0.19 |
| Tertile 1 (0.05–1.3) | ref | – | ref | – |
| Tertile 2 (1.4–2.1) | 0.68 (0.33, 1.40) | 0.29 | 3.63 (0.91, 14.4) | 0.07 |
| Tertile 3 (2.2–11.0) | 0.76 (0.36, 1.63) | 0.48 | 1.77 (0.38, 8.17) | 0.46 |
| *PFOS* | | | | |
| $Log_2$ continuous | 0.86 (0.61, 1.21) | 0.37 | 1.31 (0.70, 2.46) | 0.39 |
| Tertile 1 (0.15–3.7) | ref | – | ref | – |
| Tertile 2 (3.8–5.8) | 1.39 (0.71, 2.75) | 0.33 | 3.74 (0.97, 14.3) | 0.06 |
| Tertile 3 (5.9–36.0) | 0.94 (0.45, 1.97) | 0.88 | 2.61 (0.66, 10.5) | 0.17 |
| *PFHxS* | | | | |
| $Log_2$ continuous | 0.98 (0.76, 1.26) | 0.86 | 1.35 (0.93, 1.95) | 0.11 |
| Tertile 1 (0.10–0.77) | ref | – | ref | – |
| Tertile 2 (0.78–1.3) | 0.85 (0.42, 1.70) | 0.65 | 2.16 (0.42, 11.0) | 0.35 |
| Tertile 3 (1.4–40.0) | 1.03 (0.51, 2.10) | 0.93 | 4.90 (1.02, 22.3) | 0.04 |

OR – odds ratios; 95% CI – 95% confidence intervals.
Odds ratios for continuous variables represent a per $log_2$ increase in perfluoroalkyl substance plasma concentration.
Models are adjusted for maternal age, education, smoking status, pre-pregnancy BMI, and parity.

analyzed by tertile. In contrast, among women carrying female fetuses, effect estimates from nearly all models for PFOA and PFOS were below 1 (i.e., protective), but were not statistically significant. In the full sample, $log_2$ PFAS-fetal sex interaction terms (reference = women carrying male fetuses) provide statistical support for fetal sex-specific effects of PFOS (OR = 0.59, 95%CI: 0.37, 0.92) and PFHxS (OR = 0.73, 95%CI: 0.53, 0.99), but not PFOA (OR = 0.67, 95%CI: 0.44, 1.04).

For models examining associations with preeclampsia, effect estimates were of similar magnitude when examined separately by the sex of the fetus, but were not statistically significant in most models. However, women carrying female fetuses who were in the highest PFHxS tertile had higher odds of developing preeclampsia relative to those in the lowest tertile (OR = 4.90; 95% CI: 1.02, 22.3). In the full sample, $log_2$ PFAS-fetal sex interaction terms (reference = women carrying male fetuses) did not provide statistical support for fetal sex-specific effects of PFOA (OR = 1.04; 95%CI: 0.48, 2.24), PFOS (OR = 1.03; 95%CI: 0.48, 2.21), or PFHxS (OR = 0.99; 95%CI: 0.63, 1.62).

In linear mixed models, each doubling of concentration of PFOA or PFHxS was associated with small increases in both SBP and DBP throughout pregnancy (Table 2a); PFOS was associated with DBP, but not SBP. Associations were consistently strongest upon admission for delivery for all PFAS and SBP, as well as for PFOA and DBP. There were few temporal trends; a monotonic increase in magnitude of association was observed only for PFOA and DBP. There was no evidence of non-linearity in nearly all models, except for the association between PFHxS and DBP during the 2nd trimester (p-value for non-linear association = 0.006). As shown in Fig. 1, this association was strongest between the lower values of PFHxS and the 75th percentile, and was attenuated between the 75th and 95th percentiles.

Few discrepant findings were observed for analyses of SBP and DBP when stratified by fetal sex (Tables 2b and 2c). Effect estimates were similarly small between groups (most < 1 mmHg) and analyses in the full sample using $Log_2$ PFAS-fetal sex interaction terms generally did not provide support for sex-specific effects (p-values ranged from 0.05 to 0.85). In linear mixed models, among women carrying male fetuses, each doubling of concentration of PFOA and PFOS was associated with small increases in DBP only, but statistically significant effects were not observed for PFHxS or between all PFAS and SBP. In contrast, among

**Table 2a**
Association between first trimester $log_2$ plasma concentrations of PFOA, PFOS, and PFHxS and per-mmHg changes in systolic and diastolic blood pressure throughout pregnancy (n = 1708).

| | Systolic blood pressure (mmHg) | | Diastolic blood pressure (mmHg) | |
|---|---|---|---|---|
| | β (95% CI) | p-value | β (95% CI) | p-value |
| *PFOA* | | | | |
| Overall | 0.82 (0.23, 1.42) | 0.006 | 0.64 (0.24, 1.05) | 0.002 |
| T1 | 0.47 (−0.16, 1.10) | 0.14 | 0.53 (0.05, 1.01) | 0.03 |
| T2 | 0.35 (−0.30, 1.01) | 0.29 | 0.72 (0.24, 1.19) | 0.003 |
| T3 | 0.82 (0.13, 1.51) | 0.02 | 0.82 (0.31, 1.34) | 0.002 |
| Delivery | 1.52 (0.54, 2.50) | 0.002 | 1.11 (0.44, 1.78) | 0.001 |
| *PFOS* | | | | |
| Overall | 0.002 (−0.46, 0.48) | 0.99 | 0.47 (0.10, 0.85) | 0.01 |
| T1 | −0.12 (−0.70, 0.46) | 0.69 | 0.46 (0.01, 0.90) | 0.04 |
| T2 | −0.19 (−0.79, 0.40) | 0.53 | 0.33 (−0.10, 0.76) | 0.13 |
| T3 | 0.28 (−0.36, 0.92) | 0.40 | 0.66 (0.18, 1.14) | 0.007 |
| Delivery | 1.19 (0.28, 2.10) | 0.01 | 0.46 (−0.17, 1.08) | 0.15 |
| *PFHxS* | | | | |
| Overall | 0.63 (0.29, 0.97) | 0.0003 | 0.42 (0.16, 0.67) | 0.002 |
| T1 | 0.59 (0.18, 1.01) | 0.005 | 0.31 (−0.01, 0.62) | 0.06 |
| T2 | 0.58 (0.16, 1.00) | 0.007 | 0.52 (0.22, 0.82) | 0.001 |
| T3 | 0.72 (0.27, 1.17) | 0.002 | 0.61 (0.26, 0.95) | 0.001 |
| Delivery | 0.90 (0.25, 1.54) | 0.006 | 0.40 (−0.04, 0.84) | 0.07 |

β – parameter estimate; 95% CI – 95% confidence intervals; T1-T3 – trimesters 1 to 3.
Overall parameter estimates are derived from linear mixed models with AR(1) covariance structure including a random effect of clustering at the participant level. Trimester-specific parameter estimates are derived from general linear models. All models are adjusted for maternal age, education, smoking status, pre-pregnancy BMI, parity. Delivery refers to the blood pressure measure taken upon admission for, but prior to, delivery. Participants who reported using antihypertensive medications during pregnancy (n = 31) were excluded.

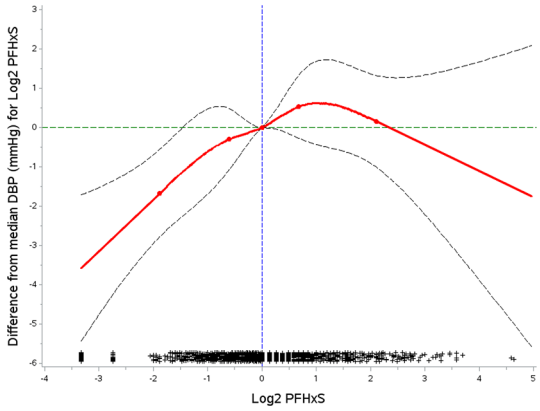


**Fig. 1.** Adjusted dose–response model of the association between 1st trimester $Log_2$ perfluorohexane sulfonate (PFHxS) plasma concentrations and 2nd trimester diastolic blood pressure (DBP, mmHg) in a cohort of Canadian pregnant women (n = 1708 after removing 31 participants taking antihypertensive medications). Restricted cubic spline functions have knots (denoted with dots) at the 5th, 25th, 50th, 75th, and 95th percentiles. The vertical dashed line (blue) denotes the median value of PFHxS and the horizontal dashed line (green) denotes the referent of DBP at the median value of PFHxS. Black dashed lines denote 95% confidence intervals. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

women carrying female fetuses, PFOA was associated with SBP and DBP upon admission for delivery only, but with larger effect sizes than other estimates. PFHxS was associated with increases in both SBP and DBP in nearly all models (with a stronger effect for SBP upon admission for delivery), but statistically significant effects were not observed for PFOS. There was no evidence of non-linearity in all models stratified by

**Table 2b**
Association between first trimester $log_2$ plasma concentrations of PFOA, PFOS, and PFHxS and per-mmHg changes in systolic and diastolic blood pressure throughout pregnancy among women carrying male fetuses (n = 897).

| | Systolic blood pressure (mmHg) | | Diastolic blood pressure (mmHg) | |
|---|---|---|---|---|
| | β (95% CI) | p-value | β (95% CI) | p-value |
| *PFOA* | | | | |
| Overall | 0.54 (−0.30, 1.37) | 0.21 | 0.68 (0.12, 1.23) | 0.02 |
| T1 | 0.32 (−0.55, 1.19) | 0.47 | 0.50 (−0.18, 1.17) | 0.15 |
| T2 | 0.29 (−0.61, 1.18) | 0.53 | 0.79 (0.15, 1.43) | 0.02 |
| T3 | 0.70 (−0.27, 1.66) | 0.16 | 0.86 (0.14, 1.59) | 0.02 |
| Delivery | 0.82 (−0.57, 2.22) | 0.25 | 0.98 (0.09, 1.87) | 0.03 |
| *PFOS* | | | | |
| Overall | −0.07 (−0.72, 0.58) | 0.84 | 0.64 (0.14, 1.14) | 0.01 |
| T1 | −0.26 (−1.06, 0.52) | 0.51 | 0.64 (0.03, 1.26) | 0.04 |
| T2 | −0.21 (−1.03, 0.59) | 0.60 | 0.50 (−0.07, 1.08) | 0.09 |
| T3 | 0.30 (−0.59, 1.19) | 0.51 | 0.68 (0.02, 1.35) | 0.04 |
| Delivery | 0.96 (−0.30, 2.23) | 0.14 | 0.50 (−0.32, 1.31) | 0.23 |
| *PFHxS* | | | | |
| Overall | 0.29 (−0.18, 0.76) | 0.22 | 0.22 (−0.13, 0.58) | 0.22 |
| T1 | 0.22 (−0.35, 0.79) | 0.44 | 0.04 (−0.40, 0.48) | 0.86 |
| T2 | 0.30 (−0.28, 0.87) | 0.31 | 0.47 (0.06, 0.88) | 0.03 |
| T3 | 0.48 (−0.14, 1.11) | 0.13 | 0.32 (−0.15, 0.79) | 0.18 |
| Delivery | 0.42 (−0.48, 1.32) | 0.36 | 0.35 (−0.23, 0.92) | 0.24 |

β – parameter estimate; 95% CI – 95% confidence intervals; T1-T3 – trimesters 1 to 3.
Overall parameter estimates are derived from linear mixed models with AR(1) covariance structure including a random effect of clustering at the participant level. Trimester-specific parameter estimates are derived from general linear models. All models are adjusted for maternal age, education, smoking status, pre-pregnancy BMI, parity. Delivery refers to the blood pressure measure taken upon admission for, but prior to, delivery. Participants who reported using antihypertensive medications during pregnancy (n = 31) were excluded.

**Table 2c**
Association between first trimester $log_2$ plasma concentrations of PFOA, PFOS, and PFHxS and per-mmHg changes in systolic and diastolic blood pressure throughout pregnancy among women carrying female fetuses (n = 809).

| | Systolic blood pressure (mmHg) | | Diastolic blood pressure (mmHg) | |
|---|---|---|---|---|
| | β (95% CI) | p-value | β (95% CI) | p-value |
| *PFOA* | | | | |
| Overall | 1.04 (0.19, 1.89) | 0.02 | 0.48 (−0.12, 1.07) | 0.12 |
| T1 | 0.53 (−0.38, 1.46) | 0.29 | 0.42 (−0.28, 1.11) | 0.24 |
| T2 | 0.47 (−0.50, 1.44) | 0.34 | 0.57 (−0.14, 1.28) | 0.12 |
| T3 | 0.79 (−0.20, 1.77) | 0.12 | 0.66 (−0.08, 1.41) | 0.08 |
| Delivery | 2.11 (0.72, 3.50) | 0.003 | 1.23 (0.22, 2.25) | 0.02 |
| *PFOS* | | | | |
| Overall | 0.01 (−0.69, 0.72) | 0.97 | 0.20 (−0.36, 0.77) | 0.48 |
| T1 | −0.06 (−0.92, 0.81) | 0.90 | 0.12 (−0.52, 0.78) | 0.70 |
| T2 | −0.11 (−0.99, 0.78) | 0.82 | 0.13 (−0.52, 0.77) | 0.70 |
| T3 | 0.09 (−0.84, 1.01) | 0.85 | 0.54 (−0.16, 1.24) | 0.13 |
| Delivery | 1.31 (−0.01, 2.63) | 0.05 | 0.38 (−0.59, 1.34) | 0.44 |
| *PFHxS* | | | | |
| Overall | 0.97 (0.47, 1.47) | 0.0001 | 0.59 (0.19, 0.99) | 0.004 |
| T1 | 0.94 (0.34, 1.55) | 0.002 | 0.52 (0.06, 0.97) | 0.03 |
| T2 | 0.92 (0.29, 1.55) | 0.004 | 0.58 (0.12, 1.03) | 0.01 |
| T3 | 0.90 (0.23, 1.56) | 0.008 | 0.88 (0.38, 1.38) | 0.0006 |
| Delivery | 1.36 (0.43, 2.30) | 0.004 | 0.45 (−0.23, 1.14) | 0.19 |

β – parameter estimate; 95% CI – 95% confidence intervals; T1-T3 – trimesters 1 to 3.
Overall parameter estimates are derived from linear mixed models with AR(1) covariance structure including a random effect of clustering at the participant level. Trimester-specific parameter estimates are derived from general linear models. All models are adjusted for maternal age, education, smoking status, pre-pregnancy BMI, parity. Delivery refers to the blood pressure measure taken upon admission for, but prior to, delivery. Participants who reported using antihypertensive medications during pregnancy (n = 31) were excluded.

sex (all p > 0.05). However, sex-stratified restricted cubic spline results for PFHxS and DBP in the 2nd trimester are presented in Figs. S1 and S2 for consistency with the full sample. The shape of the curve among women carrying female fetuses had a more pronounced association between the lower and 50th percentiles, similar to that of the full sample, but with significant attenuation between the 50th and 95th percentiles. The shape of the curve among women carrying male fetuses was non-monotonic with similar associations at the 25th, 50th, and 95th percentiles and with the strongest association at the 75th percentile.

## 4. Discussion

In this paper, we investigated associations between PFOA, PFOS, and PFHxS, and hypertensive disorders of pregnancy in women participating in a Canadian pregnancy cohort study. The prevalence of these disorders in this cohort is higher than national estimates (gestational hypertension: 7.3% in MIREC vs. 4.6% nationally; preeclampsia: 2.8% in MIREC vs. 1.1% nationally) (Public Health Agency of Canada, 2014), despite the low-risk profile of this sample (e.g., 56% multiparous, lower BMI, exclusion of mothers < 18 years old). This difference may be because we obtained information directly from medical charts in a research/clinical setting vs. using ICD classifications frequently used for administrative health databases. We found that PFOA and PFOS were not associated with the development of hypertensive disorders of pregnancy. These findings are in line with some previous reports from the literature (Huang et al., 2019; Nolan et al., 2010; Savitz et al., 2012b; Starling et al., 2014), but not all (Darrow et al., 2013; Savitz et al., 2012a; Stein et al., 2009; Wikström et al., 2019b). In the current paper we used first trimester plasma concentrations of PFAS to estimate exposure, as well as clinical measures and diagnoses to assign outcome status, which is a stronger approach compared with the recalled or predicted exposure/outcome data used in many of these previous papers. We showed that PFHxS was positively associated with the development of preeclampsia. In particular, we observed that the odds of developing preeclampsia were 3-fold higher among those in the highest (relative to lowest) tertile of PFHxS, after adjusting for relevant confounding factors. To our knowledge, this is the strongest such effect for any PFAS observed to date. It is worth noting that most other odds ratios, while not statistically significant, were in the positive direction (i.e., > 1). In addition, higher plasma concentrations of all three PFAS were associated with increases in DBP throughout pregnancy, and PFOA and PFHxS were also similarly associated with SBP. While these results were statistically significant, the magnitude of these associations is small – the largest effect observed was a 1.52 mmHg average increase in DBP for each doubling of PFOA plasma concentration. The clinical significance of these findings is likely low, but at a population level this could slightly shift the distribution of blood pressure towards an increased incidence of gestational hypertension.

Previous epidemiological studies of general obstetrical populations examining PFAS exposure have demonstrated mostly null or conflicting results. Starling et al. (2014) observed a negative association between mid-pregnancy plasma levels of perfluoroundecanoic acid and risk of developing preeclampsia, but did not observe an association for several others PFAS, including PFHxS (Starling et al., 2014). Similarly, Huang et al. (2019) primarily observed associations with perfluorobutane sulfonate, and did not observe associations for cord blood concentrations of PFHxS and gestational hypertension or preeclampsia (individually or when combined) in single-exposure models. However, in a multi-exposure model using elastic net regression these authors identified PFHxS as being weakly negatively associated with preeclampsia (but not gestational hypertension). As noted in the review by Bach et al. (2015), a direct comparison of results between studies is complicated when PFAS are measured at different time points during or around pregnancy. For instance, the first trimester plasma concentrations of PFHxS were higher in this sample of Canadian women (medians

ranging from 1.0 to 1.5 μg/L) than in the mid-pregnancy plasma samples of Norwegian women in the paper by Starling et al. (2014) (median = 0.69 ng/ml [equivalent to μg/L]) or the cord blood concentrations of Chinese women in the paper by Huang et al. (2019) (median = 0.16 ng/ml). This is not surprising because PFAS levels in maternal blood are believed to decrease throughout pregnancy (Glynn et al., 2012) which is likely related to fetal transfer, but also to dilution as a result of increases in maternal blood volume during pregnancy (Bach et al., 2015). Additionally, PFAS may adversely affect both liver (Fisher et al., 2013; Lin et al., 2010) and renal (Jain and Ducatman, 2019; Pérez et al., 2013; Stanifer et al., 2018) function, which could impact the toxicokinetics and clearance of PFAS throughout pregnancy and introduce confounding for values obtained in later pregnancy. Findings using cord blood concentrations are further influenced by issues of temporality and transplacental transfer efficiency (Wang et al., 2019).

In contrast, the first trimester plasma concentrations of PFAS collected by Wikström et al. (2019a,b) are similar to those in the current study (median PFHxS = 1.2, PFOA = 1.6, PFOS = 5.4). However, these authors showed that PFOS and perfluorononanoic acid, but not PFHxS or other PFAS, were associated with higher risk of developing preeclampsia. Differences between this and the current study with respect to exposure and confounder estimation are small (e.g., adjusting for pre-pregnancy body weight vs. BMI), and data were collected around the same time. The most notable difference is the use of different definitions of preeclampsia. In these previous studies, preeclampsia was defined as the presence of elevated blood pressure (SBP ≥ 140 mmHg and and/or DBP ≥ 90 mmHg) and proteinuria (Huang et al., 2019; Starling et al., 2014; Wikström et al., 2019b), consistent with guidelines from the American College of Obstetricians and Gynecologists (American College of Obstetricians and Gynecologists, 2002). In the present analysis, preeclampsia was defined using guidelines from the Society of Obstetricians and Gynaecologists of Canada (Magee et al., 2014), which additionally includes the presence of other maternal complications. In this study, around one-quarter (13 out of 49) of the women diagnosed with preeclampsia using this definition did not have proteinuria, and 9 of these women had elevated liver enzymes. In the paper by Wikström et al. (2019a,b), none of the 64 women diagnosed with preeclampsia displayed signs of eclampsia or HELLP-syndrome (i.e., Hemolysis, Elevated Liver enzymes, Low platelet count Syndrome). When we restricted the definition of preeclampsia to include only those women with proteinuria, effect estimates were attenuated and were not statistically significant for all three PFAS. Collectively, these findings may indicate that PFHxS is more strongly linked to maternal complications implicated in the development of preeclampsia, such as altered liver function, rather than with elevated blood pressure or proteinuria. Evidence from general adult populations provides some support for this interpretation. Most notably, in a nationally representative sample of adult Canadians sampled around the same time as the MIREC cohort, plasma levels of PFHxS, but not PFOA or PFOS, were associated with deleterious results for plasma lipids and cholesterol (Fisher et al., 2013). It is not clear why we did not observe an association with PFOS, as seen by Wikström et al. (2019a,b). Preeclampsia is a heterogeneous condition that is still poorly understood (Steegers et al., 2010), and it is possible that the participants diagnosed with preeclampsia in these two studies were different, especially given the use of different definitions of preeclampsia.

The biological mechanisms underlying associations between PFASs and preeclampsia are not well established and it is not known at what point PFAS might be involved in the progression of this disease. There is evidence, albeit limited, to suggest that PFAS can interfere with cellular lipid patterns in human placental cytotrophoblast cells (Gorrochategui et al., 2014). It is therefore plausible that some PFAS contribute to defective spiral artery remodelling by interfering with cytotrophoblast invasion and contributing to the development of the first (i.e., placental) stage of preeclampsia. Animal studies suggest that PFAS can interfere with lipid homeostasis and fatty acid metabolism, although primarily in liver cells (Andersen et al., 2008; Butenhoff et al., 2009; Kennedy et al., 2004; Lau et al., 2006), and that these effects are mediated through activation of PPARs (Andersen et al., 2008; Kennedy et al., 2004). However, the relevance of PPAR-dependent pathways for PFAS in humans is unclear (Chappell et al., 2020; Lau et al., 2007; Rosen et al., 2017; Sundström et al., 2012). This is mainly because of reduced expression of PPARs in human tissues relative to other animals (Rosen et al., 2017), but also because of interspecies differences in clearance rates and much shorter half-lives of PFAS (i.e., a few months) in non-human mammals (Lau et al., 2007; Sundström et al., 2012) vs. humans (i.e,. several years) (Olsen et al., 2007). Further research is required to elucidate relevant mechanisms in humans.

To our knowledge, this is the first paper to stratify analyses of PFAS and hypertensive disorders of pregnancy by fetal sex and these findings are entirely novel. We showed that PFOS and PFHxS were associated with gestational hypertension among women carrying male fetuses, but that effect estimates (although not statistically significant) were in the opposite direction for women carrying female fetuses. For preeclampsia, the potential for effect modification by fetal sex was generally not supported by differences in the magnitude and direction of effect estimates, but we did observe a positive, and statistically significant, effect for PFHxS (highest vs. lowest tertile) among women carrying female fetuses only. However, the confidence intervals for this association are extremely wide, which could be indicative of reduced statistical power following stratification. Collectively, these results may help to partially explain some of the null findings observed in other studies that have not been stratified by fetal sex; but, these findings should be considered exploratory because of the low number of participants following stratification. Ultimately, replication work will be needed to better understand the stability of these potential stratified effects across other populations. There is some toxicological evidence to support the potential for fetal-sex specific effects of prenatal exposure to PFAS through the inhibition of aromatase activity within human placental trophoblastic cells (Gorrochategui et al., 2014), but further toxicological evidence is also required to better understand any potential fetal sex-specific effects. While some discrepant findings were observed in sex-stratified analyses of PFAS and blood pressure, the magnitude and direction of associations was similar between women carrying male vs. female fetuses and results from the full sample using interaction terms generally did not support the potential for sex-stratified effects.

There are some relevant strengths of this study that are worth noting. MIREC is a prospective, multi-site, pan-Canadian pregnancy cohort study which is uniquely positioned to study the impact of environmental chemicals on maternal and infant/child health. Unlike many previous studies, we used measured values of PFAS and clinical assessments of hypertensive disorders of pregnancy, which reduces the likelihood of information bias. Since the genesis of preeclampsia is thought to be in the first and second trimesters (Steegers et al., 2010), the use of first trimester plasma samples is a strength of this paper. Moreover, we examined the potential effects of PFAS separately for gestational hypertension and preeclampsia, which are two separate diseases with different risk factor profiles and potentially different underlying mechanisms (Shen et al., 2017), and this helps to disentangle previously identified associations that combined these two. Nonetheless, there are important limitations to consider. First, we only measured the three PFAS that were commonly assessed at the time of cohort recruitment. There is emerging evidence that other PFAS, including those with different isomeric structures and chain lengths (Liu et al., 2019), as well as mixtures of PFASs (Hu et al., 2018), are important to consider. Second, this sample included only a relatively small number of women who developed preeclampsia. As a result, we were unable to distinguish between early (i.e., requiring delivery before 32–34 weeks of gestation) vs. late onset of preeclampsia, which is relevant because it is thought that these may actually be two separate conditions (Shen et al., 2017) in addition to being separate from gestational hypertension. However, since early preeclampsia represents a

small proportion of those who develop preeclampsia overall, failure to separate the outcome into these groups would not be expected to have substantially impacted the results observed in this cohort. With larger sample sizes, future studies could consider stratifying preeclampsia in this way to examine the potential for differential effects. Third, the high socio-economic status of this sample of pregnant Canadian women may limit the generalizability of our findings, particularly among more socio-economically disadvantaged populations. Moreover, this sample of Canadian women was predominately Caucasian and we lacked the necessary heterogeneity to include ethnicity in our analyses. This could impact the external, but not internal, validity of the current findings.

## 5. Conclusions

In this cohort of pregnant Canadian women, higher levels of PFOA were not associated with the development of hypertensive disorders of pregnancy in the full sample or in sex-stratified analyses. Higher levels of PFOS were also not associated with the development of hypertensive disorders of pregnancy in the full sample; however, among women carrying male fetuses, each doubling of concentration of PFOS was associated with higher odds of developing gestational hypertension, but not preeclampsia. PFHxS was not associated with the development of gestational hypertension, but higher levels of PFHxS were associated with the development of preeclampsia, and this effect was stronger among women carrying female (vs. male) fetuses. Associations between all three PFAS and measures of blood pressure were generally positive but with small effect sizes.

## CRediT authorship contribution statement

**Michael M. Borghese:** Conceptualization, Methodology, Formal analysis, Writing - original draft, Writing - review & editing. **Mark Walker:** Project administration, Writing - review & editing. **Michael E. Helewa:** Project administration, Writing - review & editing. **William D. Fraser:** Project administration, Funding acquisition, Methodology, Writing - review & editing. **Tye E. Arbuckle:** Project administration, Funding acquisition, Methodology, Writing - review & editing.

## Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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## Declaration of Competing Interest

All authors declare they have no potential conflicts of interest including any relevant financial interests, activities, relationships or affiliations.

## Appendix A. Supplementary material

Supplementary data to this article can be found online at https://doi.org/10.1016/j.envint.2020.105789.

## References

American College of Obstetricians and Gynecologists, 2002. Diagnosis and management of preeclampsia and eclampsia. Int. J. Gynaecol. Obstet. 77, 67–75.

Andersen, C.S., Fei, C., Gamborg, M., Nohr, E.A., Sørensen, T.I.A., Olsen, J., 2010. Prenatal exposures to perfluorinated chemicals and anthropometric measures in infancy. Am. J. Epidemiol. 172, 1230–1237. https://doi.org/10.1093/aje/kwq289.

Andersen, M.E., Butenhoff, J.L., Chang, S.-C., Farrar, D.G., Kennedy, G.L., Lau, C., Olsen, G.W., Seed, J., Wallace, K.B., 2008. Perfluoroalkyl acids and related chemistries—toxicokinetics and modes of action. Toxicol. Sci. 102, 3–14. https://doi.org/10.1093/toxsci/kfm270.

Arbuckle, T.E., Fraser, W.D., Fisher, M., Davis, K., Liang, C.L., Lupien, N., Bastien, S., Velez, M.P., von Dadelszen, P., Hemmings, D.G., Wang, J., Helewa, M., Taback, S., Sermer, M., Foster, W., Ross, G., Fredette, P., Smith, G., Walker, M., Shear, R., Dodds, L., Ettinger, A.S., Weber, J.-P., D'Amour, M., Legrand, M., Kumarathasan, P., Vincent, R., Luo, Z.-C., Platt, R.W., Mitchell, G., Hidiroglou, N., Cockell, K., Villeneuve, M., Rawn, D.F.K., Dabeka, R., Cao, X.-L., Becalski, A., Ratnayake, N., Bondy, G., Jin, X., Wang, Z., Tittlemier, S., Julien, P., Avard, D., Weiler, H., LeBlanc, A., Muckle, G., Boivin, M., Dionne, G., Ayotte, P., Lanphear, B., Séguin, J.R., Saint-Amour, D., Dewailly, É., Monnier, P., Koren, G., Ouellet, E., 2013. Cohort profile: the maternal-infant research on environmental chemicals research platform. Paediatr. Perinat. Epidemiol. 27, 415–425. https://doi.org/10.1111/ppe.12061.

Ashley-Martin, J., Dodds, L., Arbuckle, T.E., Morisset, A.S., Fisher, M., Bouchard, M.F., Shapiro, G.D., Ettinger, A.S., Monnier, P., Dallaire, R., Taback, S., Fraser, W., 2016. Maternal and neonatal levels of perfluoroalkyl substances in relation to gestational weight gain. Int. J. Environ. Res. Public Health 13. https://doi.org/10.3390/ijerph13010146.

Bach, C.C., Bech, B.H., Brix, N., Nohr, E.A., Bonde, J.P.E., Henriksen, T.B., 2015. Perfluoroalkyl and polyfluoroalkyl substances and human fetal growth: A systematic review. Crit. Rev. Toxicol. 45, 53–67. https://doi.org/10.3109/10408444.2014.952400.

Beesoon, S., Genuis, S.J., Benskin, J.P., Martin, J.W., 2012. Exceptionally high serum concentrations of perfluorohexanesulfonate in a Canadian family are linked to home carpet treatment applications. Environ. Sci. Technol. 46, 12960–12967. https://doi.org/10.1021/es3034654.

Berg, V., Nøst, T.H., Huber, S., Rylander, C., Hansen, S., Veyhe, A.S., Fuskevåg, O.M., Odland, J.Ø., Sandanger, T.M., 2014. Maternal serum concentrations of per- and polyfluoroalkyl substances and their predictors in years with reduced production and use. Environ. Int. 69, 58–66. https://doi.org/10.1016/j.envint.2014.04.010.

Brantsæter, A.L., Whitworth, K.W., Ydersbond, T.A., Haug, L.S., Haugen, M., Knutsen, H.K., Thomsen, C., Meltzer, H.M., Becher, G., Sabaredzovic, A., Hoppin, J.A., Eggesbø, M., Longnecker, M.P., 2013. Determinants of plasma concentrations of perfluoroalkyl substances in pregnant Norwegian women. Environ. Int. 54, 74–84. https://doi.org/10.1016/j.envint.2012.12.014.

Buck, R.C., Franklin, J., Berger, U., Conder, J.M., Cousins, I.T., de Voogt, P., Jensen, A.A., Kannan, K., Mabury, S.A., van Leeuwen, S.P.J., 2011. Perfluoroalkyl and polyfluoroalkyl substances in the environment: terminology, classification, and origins. Integr. Environ. Assess. Manag. 7, 513–541. https://doi.org/10.1002/ieam.258.

Butalia, S., Audibert, F., Côt, A.-M., Firoz, T., Logan, A.G., Magee, L.A., Mundle, W., Rey, E., Rabi, D.M., Daskalopoulou, S.S., Nerenberg, K.A., Canada, H., 2018. Hypertension Canada's 2018 guidelines for the management of hypertension in pregnancy. Can. J. Cardiol. 34, 526–531. https://doi.org/10.1016/j.cjca.2018.02.021.

Butenhoff, J.L., Chang, S.-C., Ehresman, D.J., York, R.G., 2009. Evaluation of potential reproductive and developmental toxicity of potassium perfluorohexanesulfonate in Sprague Dawley rats. Reprod. Toxicol. 27, 331–341. https://doi.org/10.1016/J.REPROTOX.2009.01.004.

Calafat, A.M., Wong, L.-Y., Kuklenyik, Z., Reidy, J.A., Needham, L.L., 2007. Polyfluoroalkyl chemicals in the U.S. population: data from the national health and nutrition examination survey (NHANES) 2003–2004 and comparisons with NHANES 1999–2000. Environ. Health Perspect. 115, 1596–1602. https://doi.org/10.1289/ehp.10598.

Chappell, G.A., Thompson, C.M., Wolf, J.C., Cullen, J.M., Klaunig, J.E., Haws, L.C., 2020. Assessment of the mode of action underlying the effects of GenX in mouse liver and implications for assessing human health risks. Toxicol. Pathol. https://doi.org/10.1177/0192623320905803. 019262332090580.

Darrow, L.A., Stein, C.R., Steenland, K., 2013. Serum perfluorooctanoic acid and perfluorooctane sulfonate concentrations in relation to birth outcomes in the Mid-Ohio Valley, 2005–2010. Environ. Health Perspect. 121, 1207–1213. https://doi.org/10.1289/ehp.1206372.

Desquilbet, L., Mariotti, F., 2010. Dose-response analyses using restricted cubic spline functions in public health research. Statist. Med. https://doi.org/10.1002/sim.3841. n/a–n/a.

DiGiuseppe, D.L., Aron, D.C., Ranbom, L., Harper, D.L., Rosenthal, G.E., 2002. Reliability of birth certificate data: a multi-hospital comparison to medical records information. Matern. Child Health J. 6, 169–179. https://doi.org/10.1023/A:1019726112597.

Fei, C., McLaughlin, J.K., Tarone, R.E., Olsen, J., 2007. Perfluorinated chemicals and fetal growth: a study within the Danish national birth cohort. Environ. Health Perspect. 115, 1677–1682. https://doi.org/10.1289/ehp.10506.

Fisher, M., Arbuckle, T.E., Liang, C.L., LeBlanc, A., Gaudreau, E., Foster, W.G., Haines, D.,

Davis, K., Fraser, W.D., 2016. Concentrations of persistent organic pollutants in maternal and cord blood from the maternal-infant research on environmental chemicals (MIREC) cohort study. Environ. Heal. 15, 59. https://doi.org/10.1186/s12940-016-0143-y.

Fisher, M., Arbuckle, T.E., Wade, M., Haines, D.A., 2013. Do perfluoroalkyl substances affect metabolic function and plasma lipids?—Analysis of the 2007–2009, Canadian Health Measures Survey (CHMS) Cycle 1. Environ. Res. 121, 95–103. https://doi.org/10.1016/j.envres.2012.11.006.

Glynn, A., Berger, U., Bignert, A., Ullah, S., Aune, M., Lignell, S., Darnerud, P.O., 2012. Perfluorinated alkyl acids in blood serum from primiparous women in Sweden: Serial sampling during pregnancy and nursing, and temporal trends 1996–2010. Environ. Sci. Technol. 46, 9071–9079. https://doi.org/10.1021/es301168c.

Gorrochategui, E., Pérez-Albaladejo, E., Casas, J., Lacorte, S., Porte, C., 2014. Perfluorinated chemicals: Differential toxicity, inhibition of aromatase activity and alteration of cellular lipids in human placental cells. Toxicol. Appl. Pharmacol. 277, 124–130. https://doi.org/10.1016/J.TAAP.2014.03.012.

Haines, D.A., Khoury, C., Saravanabhavan, G., Werry, K., Walker, M., Malowany, M., 2017. Human biomonitoring reference values derived for persistent organic pollutants in blood plasma from the Canadian Health Measures Survey 2007–2011. Int. J. Hyg. Environ. Health 220, 744–756. https://doi.org/10.1016/J.IJHEH.2017.03.004.

Harada, K., Inoue, K., Morikawa, A., Yoshinaga, T., Saito, N., Koizumi, A., 2005. Renal clearance of perfluorooctane sulfonate and perfluorooctanoate in humans and their species-specific excretion. Environ. Res. 99, 253–261. https://doi.org/10.1016/j.envres.2004.12.003.

Health Canada, 2013. Second Report on Human Biomonitoring of Environmental Chemicals in Canada. Results of the Canadian Health Measures Survey Cycle 2 (2009–2011). Ottawa, ON.

Houde, M., Martin, J.W., Letcher, R.J., Solomon, K.R., Muir, D.C.G., 2006. Biological monitoring of polyfluoroalkyl substances: a review. Environ. Sci. Technol. 40, 3463–3473. https://doi.org/10.1021/ES052580B.

Hu, X.C., Dassuncao, C., Zhang, X., Grandjean, P., Weihe, P., Webster, G.M., Nielsen, F., Sunderland, E.M., 2018. Can profiles of poly- and Perfluoroalkyl substances (PFASs) in human serum provide information on major exposure sources? Environ. Health 17, 11. https://doi.org/10.1186/s12940-018-0355-4.

Huang, R., Chen, Q., Zhang, L., Luo, K., Chen, L., Zhao, S., Feng, L., Zhang, J., 2019. Prenatal exposure to perfluoroalkyl and polyfluoroalkyl substances and the risk of hypertensive disorders of pregnancy. Environ. Health 18, 5. https://doi.org/10.1186/s12940-018-0445-3.

Huppertz, B., 2008. Placental origins of preeclampsia: challenging the current hypothesis. Hypertension (Dallas, Tex. 1979) 51, 970–975. https://doi.org/10.1161/HYPERTENSIONAHA.107.107607.

Huppertz, B., Kingdom, J., Caniggia, I., Desoye, G., Black, S., Korr, H., Kaufmann, P., 2003. Hypoxia favours necrotic versus apoptotic shedding of placental syncytiotrophoblast into the maternal circulation. Placenta 24, 181–190.

Jain, R.B., Ducatman, A., 2019. Perfluoroalkyl acids serum concentrations and their relationship to biomarkers of renal failure: Serum and urine albumin, creatinine, and albumin creatinine ratios across the spectrum of glomerular function among US adults. Environ. Res. 174, 143–151. https://doi.org/10.1016/J.ENVRES.2019.04.034.

Jelin, A., Cheng, Y., Shaffer, B., Kaimal, A., Little, S., Caughey, A., 2009. Early-onset preeclampsia and neonatal outcomes. J. Matern. Neonatal Med. 23, 1–5. https://doi.org/10.1080/14767050903168416.

Kannan, K., Corsolini, S., Falandysz, J., Fillmann, G., Kumar, K.S., Loganathan, B.G., Mohd, M.A., Olivero, J., Van Wouwe, N., Yang, J.H., Aldous, K.M., 2004. Perfluorooctanesulfonate and related fluorochemicals in human blood from several countries. Environ. Sci. Technol. 38, 4489–4495. https://doi.org/10.1021/es0493446.

Kärrman, A., Mueller, J.F., van Bavel, B., Harden, F., Toms, L.-M.L., Lindström, G., 2006. Levels of 12 perfluorinated chemicals in pooled australian serum, collected 2002–2003, in relation to age, gender, and region. Environ. Sci. Technol. 40, 3742–3748.

Kato, K., Wong, L.-Y., Jia, L.T., Kuklenyik, Z., Calafat, A.M., 2011. Trends in exposure to polyfluoroalkyl chemicals in the U.S. Population: 1999–2008. Environ. Sci. Technol. 45, 8037–8045. https://doi.org/10.1021/es1043613.

Kennedy, G.L., 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., 2004. The toxicology of perfluorooctanoate. Crit. Rev. Toxicol. 34, 351–384.

Lau, C., Anitole, K., Hodes, C., Lai, D., Pfahles-Hutchens, A., Seed, J., 2007. Perfluoroalkyl acids: a review of monitoring and toxicological findings. Toxicol. Sci. 99, 366–394. https://doi.org/10.1093/toxsci/kfm128.

Lau, C., Thibodeaux, J.R., Hanson, R.G., Narotsky, M.G., Rogers, J.M., Lindstrom, A.B., Strynar, M.J., 2006. Effects of perfluorooctanoic acid exposure during pregnancy in the mouse. Toxicol. Sci. 90, 510–518. https://doi.org/10.1093/toxsci/kfj105.

Lewin, A., Arbuckle, T.E., Fisher, M., Liang, C.L., Marro, L., Davis, K., Abdelouahab, N., Fraser, W.D., 2017. Univariate predictors of maternal concentrations of environmental chemicals: The MIREC study. Int. J. Hyg. Environ. Health 220, 77–85. https://doi.org/10.1016/j.ijheh.2017.01.001.

Lin, C.-Y., Lin, L.-Y., Chiang, C.-K., Wang, W.-J., Su, Y.-N., Hung, K.-Y., Chen, P.-C., 2010. Investigation of the associations between low-dose serum perfluorinated chemicals and liver enzymes in US adults. Am. J. Gastroenterol. 105, 1354–1363. https://doi.org/10.1038/ajg.2009.707.

Liu, X., Zhang, L., Chen, L., Li, J., Wang, Y., Wang, J., Meng, G., Chi, M., Zhao, Y., Chen, H., Wu, Y., 2019. Structure-based investigation on the association between perfluoroalkyl acids exposure and both gestational diabetes mellitus and glucose homeostasis in pregnant women. Environ. Int. 127, 85–93. https://doi.org/10.1016/j.envint.2019.03.035.

Lo, J.O., Mission, J.F., Caughey, A.B., 2013. Hypertensive disease of pregnancy and maternal mortality. Curr. Opin. Obstet. Gynecol. 25, 124–132. https://doi.org/10.1097/GCO.0b013e32835e0ef5.

Magee, L.A., Pels, A., Helewa, M., Rey, E., von Dadelszen, P., 2014. Diagnosis, evaluation, and management of the hypertensive disorders of pregnancy. Pregnancy Hypertens. Int. J. Women's Cardiovasc. Heal. 4, 105–145. https://doi.org/10.1016/J.PREGHY.2014.01.003.

Nakimuli, A., Nakubulwa, S., Kakaire, O., Osinde, M.O., Mbalinda, S.N., Kakande, N., Nabirye, R.C., Kaye, D.K., 2016. The burden of maternal morbidity and mortality attributable to hypertensive disorders in pregnancy: a prospective cohort study from Uganda. BMC Pregnancy Childbirth 16, 205. https://doi.org/10.1186/s12884-016-1001-1.

Nolan, L.A., Nolan, J.M., Shofer, F.S., Rodway, N.V., Emmett, E.A., 2010. Congenital anomalies, labor/delivery complications, maternal risk factors and their relationship with perfluorooctanoic acid (PFOA)-contaminated public drinking water. Reprod. Toxicol. 29, 147–155. https://doi.org/10.1016/j.reprotox.2009.10.012.

Olsen, G.W., Burris, J.M., Ehresman, D.J., Froehlich, J.W., Seacat, A.M., Butenhoff, J.L., Zobel, L.R., 2007. Half-life of serum elimination of perfluorooctanesulfonate, perfluorohexanesulfonate, and perfluorooctanoate in retired fluorochemical production workers. Environ. Health Perspect. 115, 1298–1305. https://doi.org/10.1289/ehp.10009.

Papadopoulou, E., Haug, L.S., Sabaredzovic, A., Eggesbø, M., Longnecker, M.P., 2015. Reliability of perfluoroalkyl substances in plasma of 100 women in two consecutive pregnancies. Environ. Res. 140, 421–429. https://doi.org/10.1016/j.envres.2015.04.022.

Pérez, F., Nadal, M., Navarro-Ortega, A., Fàbrega, F., Domingo, J.L., Barceló, D., Farré, M., 2013. Accumulation of perfluoroalkyl substances in human tissues. Environ. Int. 59, 354–362. https://doi.org/10.1016/J.ENVINT.2013.06.004.

Pijnenborg, R., Vercruysse, L., Hanssens, M., 2006. The uterine spiral arteries in human pregnancy: facts and controversies. Placenta 27, 939–958. https://doi.org/10.1016/J.PLACENTA.2005.12.006.

Public Health Agency of Canada, 2014. Maternal hypertension in Canada [WWW Document]. URL https://www.canada.ca/en/public-health/services/publications/healthy-living/maternal-hypertension-canada.html (accessed 1.29.19).

Rosen, M.B., Das, K.P., Rooney, J., Abbott, B., Lau, C., Corton, J.C., 2017. PPARα-independent transcriptional targets of perfluoroalkyl acids revealed by transcript profiling. Toxicology 387, 95–107. https://doi.org/10.1016/J.TOX.2017.05.013.

Rubin, D., 1987. Multiple Imputation for Nonresponse in Surveys. Wiley Series in Probability and Statistics. John Wiley & Sons Inc, New York.

Savitz, D.A., Stein, C.R., Bartell, S.M., Elston, B., Gong, J., Shin, H.M., Wellenius, G.A., 2012a. Perfluorooctanoic acid exposure and pregnancy outcome in a highly exposed community. Epidemiology 23, 386–392. https://doi.org/10.1097/EDE.0b013e31824cb93b.

Savitz, D.A., Stein, C.R., Elston, B., Wellenius, G.A., Bartell, S.M., Shin, H.M., Vieira, V.M., Fletcher, T., 2012b. Relationship of perfluorooctanoic acid exposure to pregnancy outcome based on birth records in the mid-Ohio valley. Environ. Health Perspect. 120, 1201–1207. https://doi.org/10.1289/ehp.1104752.

Shen, M., Smith, G.N., Rodger, M., White, R.R., Walker, M.C., Wen, S.W., 2017. Comparison of risk factors and outcomes of gestational hypertension and pre-eclampsia. PLoS One 12, e0175914. https://doi.org/10.1371/journal.pone.0175914.

Stanifer, J.W., Stapleton, H.M., Souma, T., Wittmer, A., Zhao, X., Boulware, L.E., 2018. Perfluorinated chemicals as emerging environmental threats to kidney health. Clin. J. Am. Soc. Nephrol. 13, 1479–1492. https://doi.org/10.2215/CJN.04670418.

Starling, A.P., Engel, S.M., Richardson, D.B., Baird, D.D., Haug, L.S., Stuebe, A.M., Klungsøyr, K., Harmon, Q., Becher, G., Thomsen, C., Sabaredzovic, A., Eggesbø, M., Hoppin, J.A., Travlos, G.S., Wilson, R.E., Trogstad, L.I., Magnus, P., Longnecker, M.P., 2014. Perfluoroalkyl substances during pregnancy and validated preeclampsia among nulliparous women in the Norwegian Mother and Child Cohort Study. Am. J. Epidemiol. 179, 824–833. https://doi.org/10.1093/aje/kwt432.

Steegers, E.A., von Dadelszen, P., Duvekot, J.J., Pijnenborg, R., 2010. Pre-eclampsia. Lancet 376, 631–644. https://doi.org/10.1016/S0140-6736(10)60279-6.

Stein, C.R., Savitz, D.A., Dougan, M., 2009. Serum levels of perfluorooctanoic acid and perfluorooctane sulfonate and pregnancy outcome. Am. J. Epidemiol. 170, 837–846. https://doi.org/10.1093/aje/kwp212.

Stuart, J.J., Bairey Merz, C.N., Berga, S.L., Miller, V.M., Ouyang, P., Shufelt, C.L., Steiner, M., Wenger, N.K., Rich-Edwards, J.W., 2013. Maternal recall of hypertensive disorders in pregnancy: a systematic review. J. Women's Health 22, 37–47. https://doi.org/10.1089/jwh.2012.3740.

Sundström, M., Chang, S.-C., Noker, P.E., Gorman, G.S., Hart, J.A., Ehresman, D.J., Bergman, Å., Butenhoff, J.L., 2012. Comparative pharmacokinetics of perfluorohexanesulfonate (PFHxS) in rats, mice, and monkeys. Reprod. Toxicol. 33, 441–451. https://doi.org/10.1016/j.reprotox.2011.07.004.

Trudel, D., Horowitz, L., Wormuth, M., Scheringer, M., Cousins, I.T., Hungerbühler, K., 2008. Estimating consumer exposure to PFOS and PFOA. Risk Anal. 28, 251–269. https://doi.org/10.1111/j.1539-6924.2008.01017.x.

van Esch, J.J.A., van Heijst, A.F., de Haan, A.F.J., van der Heijden, O.W.H., 2017. Early-onset preeclampsia is associated with perinatal mortality and severe neonatal morbidity. J. Matern. Neonatal Med. 30, 2789–2794. https://doi.org/10.1080/14767058.2016.1263295.

Vatten, L.J., Skjærven, R., 2004. Offspring sex and pregnancy outcome by length of gestation. Early Hum. Dev. 76, 47–54. https://doi.org/10.1016/J.EARLHUMDEV.2003.10.006.

Velez, M.P., Arbuckle, T.E., Fraser, W.D., 2015. Maternal exposure to perfluorinated chemicals and reduced fecundity: the MIREC study. Hum. Reprod. 30, 701–709. https://doi.org/10.1093/humrep/deu350.

Wallis, A.B., Saftlas, A.F., Hsia, J., Atrash, H.K., 2008. Secular trends in the rates of

preeclampsia, eclampsia, and gestational hypertension, United States, 1987–2004. Am. J. Hypertens. 21, 521–526. https://doi.org/10.1038/ajh.2008.20.
Wang, Y., Han, W., Wang, C., Zhou, Y., Shi, R., Bonefeld-Jørgensen, E.C., Yao, Q., Yuan, T., Gao, Y., Zhang, J., Tian, Y., 2019. Efficiency of maternal-fetal transfer of perfluoroalkyl and polyfluoroalkyl substances. Environ. Sci. Pollut. Res. 26, 2691–2698. https://doi.org/10.1007/s11356-018-3686-3.
White, I.R., Royston, P., Wood, A.M., 2011. Multiple imputation using chained equations: Issues and guidance for practice. Stat. Med. 30, 377–399. https://doi.org/10.1002/sim.4067.
Wikström, S., Lin, P.I., Lindh, C.H., Shu, H., Bornehag, C.G., 2019a. Maternal serum levels of perfluoroalkyl substances in early pregnancy and offspring birth weight. Pediatr. Res. 1–7. https://doi.org/10.1038/s41390-019-0720-1.
Wikström, S., Lindh, C.H., Shu, H., Bornehag, C.-G., 2019b. Early pregnancy serum levels of perfluoroalkyl substances and risk of preeclampsia in Swedish women. Sci. Rep. 9, 9179. https://doi.org/10.1038/s41598-019-45483-7.
Ying, W., Catov, J.M., Ouyang, P., 2018. Hypertensive disorders of pregnancy and future maternal cardiovascular risk. J. Am. Heart Assoc. 7, e009382. https://doi.org/10.1161/JAHA.118.009382.