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Maternal Dioxin Exposure and Pregnancy Outcomes Over 30 Years of Follow-Up in Seveso

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Abstract

Animal evidence suggests an association between exposure to 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD) and adverse pregnancy outcomes. Epidemiologic studies report inconsistent results, but are limited by narrow range of exposure, small sample size, and lack of a biologic measure of highest lifetime exposure. On July 10, 1976, a chemical explosion in Seveso, Italy resulted in the highest known residential exposure to TCDD. In 1996, we initiated the Seveso Women's Health Study (SWHS), a retrospective cohort of TCDD exposure and reproductive health. Individual-level TCDD was measured in serum collected soon after the explosion. After 20 years of follow-up, we found no association between maternal TCDD in 1976 serum or estimated at pregnancy and spontaneous abortion (SAB), fetal growth, or gestational length. Here, we present an updated analysis of TCDD exposure and adverse pregnancy outcomes from a subsequent follow-up of the SWHS cohort in 2008-2009.

SWHS women had 1,211 post-explosion pregnancies through the 2008-2009 follow-up. We found no association between TCDD estimated at pregnancy and SAB, fetal growth, or gestational length. However, we found a non-significant inverse association between maternal 1976 serum TCDD and birthweight (adjusted β=−22.8, 95% CI: −80.1, 34.6). The association was stronger among first post-explosion births, but remained non-significant (adjusted β=−47.7, 95% CI: −107.3, 11.9). SWHS is the first study to be able to consider two potentially relevant measures of TCDD exposure: highest lifetime dose and *in utero*. Our results, although non-significant, suggest that highest dose may be more relevant in epidemiologic studies of TCDD and pregnancy outcomes.

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Keywords

birthweight; dioxin; fetal growth; pregnancy outcomes; preterm; Seveso

1. INTRODUCTION[1]

In European countries, neonatal and infant mortality rates have decreased significantly over the past several decades, largely due to improved maternal and child health services, higher quality of life, and advances in neonatal care (Euro-Peristat Project with SCPE and EUROCAT 2013). Despite these drastic improvements, the principal causes of neonatal morbidity and mortality (preterm birth, restricted fetal growth, and birth defects) remain important public health problems because of the lifetime burden of disease associated with these conditions (Barouki et al. 2012; Hanson and Gluckman 2008; Hanson and Gluckman 2011; Saigal and Doyle 2008). Although several risk factors for adverse pregnancy outcomes have been established, the root causes are largely unknown; recent evidence suggests that environmental exposures could play a role (Nieuwenhuijsen et al. 2013; Slama and Cordier 2010; Stillerman et al. 2008; Windham and Fenster 2008).

The environmental contaminant 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD) is a known carcinogen and potent endocrine disruptor, and is produced as an unintentional byproduct of several industrial processes (Baan et al. 2009; Birnbaum 1995; Birnbaum 1994; IARC 1997; Wikoff et al. 2012; Zook and Rappe 1994). TCDD is the most toxic congener of a group of structurally related compounds including polychlorinated dibenzodioxins (PCDDs), dibenzofurans (PCDFs) and biphenyls (PCBs) that bind with varying strengths to the aryl hydrocarbon receptor (IARC 1997). In animal studies, maternal exposure to TCDD is associated with increased fetal and neonatal loss (Allen et al. 1979; McNulty 1984; Murray et al. 1979; Nau et al. 1986; Roman and Peterson 1998) and reduced neonatal body weight (Gray and Ostby 1995; Jin et al. 2010; Myllymaki et al. 2005; Roman and Peterson 1998; Sommer et al. 1996). However, epidemiologic studies of TCDD or total dioxin toxic equivalents (TEQ) are limited and inconsistent. Some have found associations between *in utero* TCDD or TEQ exposure and increased incidence of spontaneous abortion (SAB) (Revich et al. 2001), higher rates of urinary tract birth defects (Cordier et al. 2004; Cordier et al. 2010), and lower birthweight in male, but not female infants (Konishi et al. 2009; Vartiainen et al. 1998); others have found no association with birthweight (Halldorsson et al. 2009; Nishijo et al. 2008; Tajimi et al. 2005; Tawara et al. 2009). Limitations of these studies might account for inconsistencies: the use of exposure proxies instead of biologic measurements (Cordier et al. 2004; Cordier et al. 2010; Revich et al. 2001), inclusion of mothers with a limited range of TCDD exposure close to background levels (Halldorsson et al. 2009; Konishi et al. 2009; Nishijo et al. 2008; Tajimi et al. 2005; Tawara et al. 2009; Vartiainen et al. 1998), and small sample size (Halldorsson et al. 2009; Nishijo et al. 2008; Tawara et al. 2009; Vartiainen et al. 1998).

Studies in highly exposed populations with a wider range of exposure may help clarify the connection between TCDD exposure and pregnancy outcomes. Women exposed to dioxin-like compounds including PCDDs, PCDFs, and PCBs after a rice oil contamination incident in Japan in 1968 (Yusho) had higher rates of SAB and pregnancy loss in the decade after exposure compared to the decade before (Tsukimori et al. 2008). TCDD and TEQ levels measured in serum from 101 women collected over three decades after the Yusho incident

[1]**List of Abbreviations:** IQR=interquartile range; OR=odds ratio; PCB=polychlorinated biphenyl; PCDD=polychlorinated dibenzodioxin; PCDF=polychlorinated dibenzofuran; ppt=parts-per-trillion; SAB=spontaneous abortion; SGA=small-for-gestational age; SWHS=Seveso Women's Health Study; TEQ=toxic equivalent; TCDD=2,3,7,8-tetrachlorodibenzo-*p*-dioxin

and back-extrapolated to the time of 190 pregnancies were inversely associated with birthweight in male, but not female infants (Tsukimori et al. 2012), corroborating previous findings of effect modification by sex in background-exposed populations (Konishi et al. 2009; Vartiainen et al. 1998). However, a biologic measurement near the time of highest exposure was not available in the Yusho study.

On July 10, 1976, a chemical explosion in Seveso, Italy released up to 30 kg of TCDD over an 18 km^2 area, resulting in one of the highest residential TCDD exposures ever recorded. The surrounding area was divided into exposure zones (A, B, R, non-ABR) based on surface soil TCDD measurements (di Domenico et al. 1980). Initiated in 1996, the Seveso Women's Health Study (SWHS) is a historical cohort study of the female population residing around Seveso at the time of the explosion and represents the largest female population with known individual-level TCDD exposure (Eskenazi et al. 2000). Previously, after 20 years of follow-up, we found no association between maternal TCDD measured in 1976 serum or estimated at pregnancy and SAB, birthweight, or gestational age in 888 post-explosion pregnancies to 510 SWHS women (Eskenazi et al. 2003). However, in 1996, around 25% of SWHS women were still nulligravid, including many of the youngest, most highly exposed women (Eskenazi et al. 2004). Here, we present an updated analysis of TCDD exposure and pregnancy outcomes from a subsequent follow-up of the SWHS cohort in 2008-2009. This analysis includes 323 additional pregnancies, 64% of which are to previously nulliparous women.

2. METHODS

2.1 Study population

Details of the study design are presented elsewhere (Eskenazi et al. 2000). Briefly, eligible women were newborn to 40 years old and resided in Zones A or B on July 10, 1976, and had adequate stored serum collected soon after the explosion. Enrollment took place from 1996-1998, and 981 women (80% of eligible) participated. We conducted a follow-up of the cohort in 2008-2009, and 833 women (85%) participated.

Among 1,211 post-explosion pregnancies to 617 women, we limited the SAB analysis to 1,071 pregnancies that did not end in voluntary abortion (n=125), ectopic pregnancy (n=14), or molar pregnancy (n=1). We described birth defects among 911 pregnancies that did not end in SAB (n=160). We additionally excluded stillbirths (n=5), multiple births (n=25), and births to women with hypertension during pregnancy or gestational diabetes (n=73) for the analysis of fetal growth and gestational length. Birthweight was missing for one pregnancy, leaving a final sample of 807 infants for fetal growth, and 808 for gestational length.

2.2 Procedure

The study was approved by the Institutional Review Boards of the participating institutions, and written informed consent was obtained from all women prior to participation. Details of the study procedure for both the 1996-1998 and the 2008-2009 studies are described elsewhere (Eskenazi et al. 2000; Warner et al. 2011). At both follow-ups, personal interviews were conducted in private by a trained nurse-interviewer who was blinded to the participants' serum TCDD levels. During the interviews, women were asked a series of questions about each of their pregnancies, including pregnancy outcome, date the pregnancy ended, gestational length, weight gain, complications, and for live births, infant sex, birthweight, and presence of birth defects. Detailed medical histories and demographic information were also collected during the interviews.

2.3 Laboratory analysis

Initial TCDD was measured in archived sera collected soon after the explosion by high-resolution gas chromatography/high-resolution mass spectrometry methods for the full SWHS cohort (Patterson et al. 1987). Details of the serum sample selection and TCDD concentrations measured in 1976 serum are presented elsewhere (Eskenazi et al. 2004; Eskenazi et al. 2000). There was an inadequate volume of serum available to measure individual-level total TEQ, but initial analyses of pooled 1976 serum samples revealed that Seveso residents were highly exposed to TCDD, but not to other PCDD/Fs (Mocarelli et al. 1990).

TCDD was also measured in archived sera collected at the 1996-1998 study by high-resolution gas chromatography/isotope-dilution high-resolution mass spectrometry for 146 mothers with live births that ended after the 1996-1998 follow-up (Patterson and Turner 2005). Details of TCDD concentrations in 1996 serum are presented elsewhere (Warner et al. 2013). All values are reported on a lipid-weight basis in parts-per-trillion (ppt) (Akins et al. 1989). Non-detectable values were assigned a value of one-half the detection limit (Hornung and Reed 1990).

2.4 Statistical analysis

Statistical analyses were conducted using Stata 11.0 (Stata Corp 2009). We assessed TCDD exposure as 1) initial, highest TCDD levels (maternal 1976 serum TCDD), to test the hypothesis that initial dose produces a permanent effect on the reproductive system; and 2) estimated TCDD at pregnancy (for live birth outcomes), to test the hypothesis that direct fetal exposure results in adverse pregnancy outcomes. For pregnancies ending after July 10, 1976 through the 1996-1998 follow-up, we estimated TCDD at pregnancy from 1976 TCDD levels using methods previously described (Eskenazi et al. 2003). For new pregnancies ending after the 1996-1998 follow-up, we estimated TCDD at pregnancy from 1996 TCDD levels using a first-order kinetic model, assuming a nine year half-life (Pirkle et al. 1989).

Maternal TCDD levels in 1976 serum and estimated at pregnancy were analyzed as continuous variables ($\log_{10}$-transformed). Fetal growth was assessed continuously (birthweight controlling for gestational age), and categorically (small-for-gestational-age (SGA), defined as birthweight $<10^{th}$ percentile for gestational age and sex, based on Italian population statistics (Festini et al. 2004)). Length of gestation was assessed continuously (days) and categorically (preterm, defined as delivery prior to 37 weeks gestation). We examined all pregnancies ending between July 10, 1976 and the 2008-2009 follow-up, and first pregnancies within the same period.

We used logistic regression to examine the relationship between maternal TCDD and SAB, SGA, and preterm delivery. We used linear regression to examine the association between maternal TCDD and birthweight and gestational age. For all regression models, we used the clustered sandwich estimator of variance to allow for non-independence of multiple pregnancies to the same woman.

Potential confounders were selected *a priori* from the literature and included maternal age (years) at explosion and pregnancy, total parity, total gravidity, pre-explosion history of SAB and low birthweight, education, smoking during pregnancy, marital status at pregnancy, maternal height (cm), year of pregnancy, and infant sex. Covariates were included in the final models if they changed the coefficient on TCDD by >10%. We tested for effect modification by infant sex, using $p<0.2$ as a cut-point for significance.

In sensitivity analyses, we reran all models excluding pregnancies that were conceived prior to the explosion (n=32) or that ended before serum collection (n=42). We also reran the models for fetal growth excluding preterm births (n=44).

3. RESULTS

Table 1 presents maternal 1976 serum TCDD concentrations across categories of pregnancy and sociodemographic characteristics for 617 SWHS women and their 1,211 post-explosion pregnancies. At the time of the explosion, the women averaged 16.9 (±9.0) years of age, about one-third were premenarche, and most (74.2%) were nulliparous. Women averaged 29.9 (±5.5) years of age at time of pregnancy, and about 20% had children after age 35. Around 10% of women with live births smoked during pregnancy, and women reported an average weight gain of 12.9 kg. Of the live born infants, 52.7% were male. For the 617 women, the median maternal 1976 serum TCDD was 55.0 ppt (interquartile range (IQR)=27.0-148.0; range 2.5-56,000 ppt) and the median estimated TCDD at pregnancy (live births only) was 9.9 ppt (IQR=4.3-29.6; range=0.3-5,730 ppt). Women who were younger, premenarche, and nulliparous at explosion had significantly higher 1976 serum TCDD levels.

Among the 1,211 post-explosion pregnancies, there were 906 (74.8%) live births including 881 singletons and 25 multiple births. Non-live birth outcomes included 160 (13.2%) SABs, 14 (1.2%) ectopic pregnancies, 1 (0.1%) molar pregnancy, 125 (10.3%) voluntary abortions, and 5 (0.4%) stillbirths.

SWHS women reported 160 post-explosion SABs. Women who were older at pregnancy and had a pre-explosion history of SAB were more likely to have an SAB (data not shown). A 10-fold increase in maternal 1976 serum TCDD was not associated with SAB (adjusted odds ratio (OR)=0.78, 95% CI: 0.59, 1.02). Results were similar when the analysis was limited to first pregnancies only (n=75 SABs; adjusted OR=0.81, 95% CI: 0.55, 1.18).

Due to the small number of birth defects cases and the limitations inherent in classifying birth defects based on maternal report, we did not perform a statistical analysis for this outcome. Mothers reported one case of anencephaly, one case of cleft lip in a child who had toxoplasmosis, one case of cleft palate, and two cases of hypospadias/epispadias. Maternal 1976 serum TCDD levels for these cases were 19.5, 29.9, 150, 61.2, and 74.7 ppt, respectively.

Among the 807 live births, average birthweight was 3,278 (±498) grams. There were 44 (5.5%) low birthweight infants (<2,500 grams), and 70 (8.7%) SGA infants. Compared to infants with normal fetal growth, these infants were more likely to be female, or to have mothers who were shorter, previously nulliparous, or had a history of low birthweight (data not shown). Table 2 presents the relationship between maternal 1976 serum TCDD and fetal growth. A 10-fold increase in 1976 serum TCDD was not associated with birthweight (adjusted β=−22.8, 95% CI: −80.1, 34.6) or SGA (adjusted OR=1.19, 95% CI: 0.82, 1.74). When we limited the analysis to the first post-explosion births, point estimates were further from the null but the results remained non-significant for birthweight (adjusted β=−47.7, 95% CI: −107.3, 11.9) and SGA (adjusted OR=1.31, 95% CI: 0.86, 1.99). Limiting the analysis to term births did not change the results (adjusted β for birthweight=−26.3, 95% CI: −84.2, 31.6; adjusted OR for SGA=1.21, 95% CI: 0.83, 1.77).

Among 808 live births, the mean gestational age was 39.5 (±1.7) weeks, and 44 (5.5%) infants were preterm. Infants whose mothers had a history of low birthweight were more likely to be preterm (data not shown). Table 3 presents the relationship between maternal 1976 serum TCDD and length of gestation. A 10-fold increase in maternal 1976 serum

TCDD was not associated with gestational age (adjusted β=−0.45, 95% CI: −1.95, 1.04) or preterm delivery (adjusted OR=1.22, 95% CI: 0.73, 2.05). When we limited the analysis to first post-explosion births, the estimate was further from the null for gestational age (adjusted β=−0.63, 95% CI: −2.34, 1.09), but not for preterm delivery (adjusted OR=1.22, 95% CI: 0.69, 2.14), and results remained non-significant.

Tables 2 and 3 present the relationship between TCDD estimated at pregnancy and fetal growth and length of gestation. We found no evidence of a relationship between TCDD estimated at pregnancy and birthweight (adjusted β=13.6, 95% CI: −55.5, 82.8), SGA (adjusted OR=1.06, 95% CI: 0.65, 1.72), gestational age (adjusted β=0.17, 95% CI: −1.42, 1.77), or preterm delivery (adjusted OR=0.79, 95% CI: 0.42, 1.51). Results were similar when we limited the sample to first post-explosion births. Compared to the analyses of maternal 1976 serum TCDD, point estimates for TCDD estimated at pregnancy were closer to the null and less significant.

Among all post-explosion births, we found no evidence of effect modification by infant sex for maternal 1976 serum TCDD and birthweight (p-interaction=0.91), SGA (p-interaction=0.29), gestational age (p-interaction=0.74), or preterm delivery (p-interaction=0.69). Likewise, there was no evidence of effect modification by infant sex for TCDD at pregnancy and birthweight (p-interaction=0.96), SGA (p-interaction=0.84), gestational age (p-interaction=0.89), or preterm delivery (p-interaction=0.72). Among first post-explosion births, results were similar for birthweight, SGA, and gestational age. However, the association between maternal 1976 serum TCDD and preterm delivery was stronger in females (adjusted OR=1.85, 95% CI: 0.89, 3.84) than in males (adjusted OR=0.89, 95% CI: 0.39, 1.99) (p-interaction=0.18), although the results did not reach significance in either group. This interaction was no longer significant for the association with TCDD estimated at pregnancy (p-interaction=0.22).

Excluding pregnancies that were conceived prior to the explosion or that ended before serum collection did not appreciably alter the results.

4. DISCUSSION

To our knowledge, SWHS is the first epidemiologic study to assess the relation of TCDD measured in preconception serum and pregnancy outcomes in a highly exposed population. Consistent with our previous findings in the SWHS cohort 20 years after the explosion (Eskenazi et al. 2003), with 10 more years of follow-up and 323 additional pregnancies, we found no evidence of a relationship between maternal 1976 serum TCDD and SAB or length of gestation, and an inverse non-significant association between TCDD and birthweight. We found no relationship between TCDD estimated at pregnancy and any adverse pregnancy outcomes. The addition of mothers with new pregnancies, who were younger and more highly exposed in 1976 than the mothers reported on previously, brought our estimates for associations of 1976 TCDD and birthweight further from the null than in our previous report (−23 vs −4 grams for all births, −48 vs. −34 grams for first births); the estimates for other outcomes were similar (Eskenazi et al. 2003).

SWHS is the first study to be able to consider two potentially relevant measures of TCDD exposure: highest lifetime dose and *in utero* levels. Studies in background-exposed populations with a biologic measure of *in utero* exposure (Halldorsson et al. 2009; Konishi et al. 2009; Nishijo et al. 2008; Tajimi et al. 2005; Tawara et al. 2009; Vartiainen et al. 1998) and studies conducted in women affected by the Yusho incident (Tsukimori et al. 2012) have not considered historical exposure and do not have a biologic measure of highest lifetime dose. In SWHS, we found stronger results with maternal 1976 serum TCDD than

with TCDD estimated at pregnancy, suggesting that highest dose may be more relevant than pregnancy dose in epidemiologic studies of pregnancy outcomes. This indicates that previous studies could suffer from significant exposure misclassification. The only other studies with the ability to look at highest lifetime exposure were conducted in Seveso. They found no significant rise in incidence of SAB (from 1976-1980) or birth defects (from 1976-1982) in women from Zones A, B, and R (Fara and Del Corno 1985; Mastroiacovo et al. 1988), findings which are consistent with SWHS results. However, these studies used zone as a proxy for exposure; therefore, the estimate of initial exposure is crude.

Although we report a non-significant inverse relationship between 1976 TCDD and birth weight, we did not find an association of TCDD at pregnancy with birth weight, and no evidence of effect modification by infant sex. This is inconsistent with two epidemiologic studies that have found evidence of an association between *in utero* TCDD exposure and lower birthweight in male infants only (Konishi et al. 2009; Tsukimori et al. 2012). In a study of 398 Japanese women, TEQ was measured in maternal serum collected between 23 weeks gestation and 1 week post-delivery (Konishi et al. 2009). A 10-fold increase in *in utero* PCDD TEQ levels was associated with a significant 331.4 gram reduction in male birthweight and a non-significant 126.3 gram reduction in female birthweight. Likewise, in 101 women affected by the Yusho incident, a 10-fold increase in estimated *in utero* TCDD levels was associated with a significant 164 gram reduction in male birthweight, but a non-significant 76.3 gram reduction in female birthweight (Tsukimori et al. 2012). However, the possibility of selection bias exists in both of these studies, as participation was low (<50%) compared with SWHS (>80% for both follow-up studies). Also, in the Yusho study, *in utero* exposure measurement error is possible, as exposure was back-extrapolated to pregnancies up to 33 years earlier, and extrapolation did not consider continuous or current exposure or other factors that could influence serum levels, including pregnancy and lactation.

SWHS provides a unique opportunity to assess the relationship between maternal TCDD measured in preconception serum and pregnancy outcomes in a population highly exposed to TCDD, but not to other dioxin-like compounds. SWHS represents the largest female population with individual-level TCDD exposure data and is one of the only studies to have a biologic measure of highest lifetime TCDD exposure (Eskenazi et al. 2000). Other strengths of this study include its prospective design, high participation rate, and assessment of multiple pregnancy outcomes.

The study does have some limitations. Despite having an accurate measure of highest TCDD exposure, we do not have a biologic measure of TCDD at pregnancy, and were forced to rely on models to estimate TCDD at pregnancy. This would result in non-differential measurement error if *in utero* exposure is the relevant dose, which would bias our results towards the null. To study this possibility, a pharmacokinetic model that considers factors such as pregnancy, lactation, and growth is currently under development (Emond et al. 2012; Emond et al. 2005), and serum collected from SWHS women at both follow-ups is available for future analyses. The use of the pharmacokinetic model and the availability of subsequent TCDD measurements will enable us to better estimate exposure at the time of pregnancy.

It was also not possible to confirm pregnancy outcomes with medical records or birth certificates, and instead, we relied on maternal report (average recall=9.6 years). However, previous studies have shown that women tend to report the birthweight of their infants accurately (Rice et al. 2007), and gestational age reasonably well (Yawn et al. 1998). Further, we previously compared maternal report of birthweight with medical records for a sample of 139 SWHS births, and found that although women over-reported birthweight by about 22 grams, the bias did not vary by TCDD exposure (Eskenazi et al. 2003). Thus, we

have no reason to believe that outcome misclassification was differential with respect to exposure.

Lastly, our sample was limited to women with knowledge of a pregnancy. In SWHS, we have previously reported a longer time to pregnancy and increased odds of infertility associated with TCDD exposure (Eskenazi et al. 2010). If the most highly exposed women could not achieve or maintain pregnancy, they would not be included in our analytic sample (Eskenazi et al. 1995; Wilcox et al. 1988). Therefore, we may have underestimated the effect of TCDD exposure on birth outcomes by not including those women that may have been the most susceptible.

Although we found only limited evidence of an association between TCDD exposure and pregnancy outcomes in this cohort of postnatally-exposed women, it is possible that an effect will emerge in subsequent generations. In a study of adult female mice that were exposed to TCDD as fetuses or neonates, researchers found a near complete loss of progesterone receptor A and B expression in the uterus, resulting in decreased uterine sensitivity to progesterone (Nayyar et al. 2006), which has been associated with pregnancy loss in mice and humans (Brown et al. 2004; Druckmann and Druckmann 2005; Lydon et al. 1996; Virgo and Bellward 1974). In a subsequent study, researchers found an increased risk of infertility and preterm birth in female mice developmentally exposed to dioxin, and in three subsequent generations (Bruner-Tran and Osteen 2011). Since all SWHS women were exposed postnatally, the infants assessed in the current analysis are comparable to the developmentally-exposed generation described above, the first generation in which an effect on pregnancy outcomes was seen. Therefore, it is possible that adverse effects will emerge in the grandchildren and great-grandchildren of SWHS women. Follow-up of the second and third generations will begin shortly to test this hypothesis.

At last follow-up in 2008, about 20% of SWHS women were still of reproductive age (<40 years); thus we expect additional pregnancies will occur over the next decade. Continued follow-up of the cohort to report on all post-explosion pregnancies may be informative.

In conclusion, in this updated analysis of the SWHS cohort, we found no evidence of an association between estimated *in utero* TCDD levels and pregnancy outcomes over 30 years of follow-up. However, we found a non-significant inverse relationship between maternal 1976 TCDD levels and birthweight. Given the evidence in animals that *in utero* exposure to TCDD may affect pregnancy outcomes for several generations, continued follow-up of the cohort will be informative in assessing multi-generational effects of TCDD exposure.

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RESEARCH HIGHLIGHTS

- A 1976 factory explosion in Seveso, Italy exposed residents to high levels of TCDD.
- The Seveso Women's Health Study follows these TCDD-exposed women over 30 years.
- We examined the relation of TCDD in 1976 and at pregnancy with pregnancy outcomes.
- TCDD levels at pregnancy were not related to adverse pregnancy outcomes.
- Initial TCDD levels were non-significantly inversely related to birthweight.

Table 1

Sociodemographic and pregnancy characteristics by maternal 1976 serum TCDD levels for 617 women with 1211 post-explosion pregnancies, SWHS, Seveso, Italy, 1976-2009.

Characteristic	N (%)	1976 Serum TCDD (ppt) Median (IQR)
Total women	617 (100.0)	55.0 (27.0, 148.0)
Age at explosion (years)*		
0-10	155 (25.1)	153.0 (50.6, 327.0)
11-20	249 (40.4)	50.8 (24.3, 100.0)
21-30	164 (26.6)	38.8 (21.4, 84.0)
31-40	49 (7.9)	38.6 (27.7, 92.7)
Menarche status at explosion*		
Premenarche	202 (32.7)	125.5 (49.0, 276.0)
Postmenarche	415 (67.3)	42.1 (22.5, 88.0)
Pre-explosion parity*		
0	458 (74.2)	66.8 (29.6, 185.0)
1	81 (13.1)	36.6 (21.6, 72.0)
≥2	78 (12.6)	35.7 (25.1, 70.3)
Maternal education at last follow-up*		
<Required	146 (23.7)	40.5 (23.9, 80.9)
Required/high school	444 (72.0)	63.0 (28.7, 169.0)
University	27 (4.4)	46.8 (26.3, 192.0)
Total pregnancies	1211 (100.0)	
Age at pregnancy (years)		
<25	201 (16.6)	45.3 (21.3, 100.0)
25-29	392 (32.4)	54.7 (27.0, 126.0)
30-34	360 (29.7)	61.0 (26.0, 162.7)
≥35	258 (21.3)	48.7 (27.6, 128.0)
Smoking during pregnancy[a,*]		
No	813 (89.7)	60.2 (27.7, 156.0)
Yes	93 (10.3)	45.4 (20.7, 98.3)
Weight gain during pregnancy (kg)[a,b]		
<10	189 (21.5)	67.9 (38.2, 141.0)
10-14	422 (48.0)	52.9 (25.4, 135.0)
15-19	171 (19.4)	52.9 (22.0, 192.0)
≥20	98 (11.1)	62.2 (26.3, 126.0)
Infant sex[a]		
Male	477 (52.7)	54.2 (26.6, 126.0)
Female	429 (47.4)	63.3 (27.0, 163.4)

*Anova p-value <0.05

[a]Live-births only

[b] Missing data on pregnancy weight gain or 26 live births.

Table 2

Results of linear and logistic regression models[a] showing the relationship between maternal TCDD exposure and fetal growth, SWHS, Seveso, Italy, 1976-2009.

Exposure	Pregnancies	Birthweight		Small-for-Gestational-Age	
		n	Adj. β (95% CI)[b]	n/Total	Adj. OR (95% CI)[c]
1976 Serum TCDD	All	807	−22.8 (−80.1, 34.6)	70/807	1.19 (0.82, 1.74)
	First	518	−47.7 (−107.3, 11.9)	49/518	1.31 (0.86, 1.99)
Estimated TCDD at Pregnancy[d]	All	799	13.6 (−55.5, 82.8)	67/799	1.06 (0.65, 1.72)
	First	513	−23.2 (−96.9, 50.6)	48/513	1.18 (0.69, 2.04)

[a] Results are for a 10-fold increase in serum TCDD.

[b] Adjusted for gestational age, maternal height, pre-explosion history of low birthweight, year of pregnancy, parity, and maternal age.

[c] Adjusted for maternal height, pre-explosion history of low birthweight, year of pregnancy, parity, and maternal age.

[d] Estimated pregnancy TCDD missing for 8 births to 5 mothers.

Table 3

Results of linear and logistic regression models[a] showing the relationship between maternal TCDD exposure and length of gestation, SWHS, Seveso, Italy, 1976-2009.

Exposure	Pregnancies	Gestational Age		Preterm	
		n	Adj. β (95% CI)[b]	n/Total	Adj. OR (95% CI)[b]
1976 Serum TCDD	All	808	−0.45 (−1.95, 1.04)	44/808	1.22 (0.73, 2.05)
	First	518	−0.63 (−2.34, 1.09)	31/518	1.22 (0.69, 2.14)
Estimated TCDD at Pregnancy[c]	All	800	0.17 (−1.42, 1.77)	43/800	0.79 (0.42, 1.51)
	First	513	−0.01 (−1.81, 1.80)	30/513	0.82 (0.40, 1.68)

[a] Results are for a 10-fold increase in serum TCDD.

[b] Adjusted for maternal height, pre-explosion history of low birthweight, year of pregnancy, parity, and maternal age.

[c] Estimated pregnancy TCDD missing for 8 births to 5 mothers.