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Authors
Mannetje, Andrea T
Eng, Amanda
Walls, Chris
et al.

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ORIGINAL ARTICLE

Sex ratio of the offspring of New Zealand phenoxy herbicide producers exposed to 2,3,7,8-tetrachlorodibenzo-p-dioxin

Andrea 't Mannetje,[1] Amanda Eng,[1] Chris Walls,[2] Evan Dryson,[2] Manolis Kogevinas,[3] Collin Brooks,[1] Dave McLean,[1] Soo Cheng,[1] Allan H Smith,[4] Neil Pearce[1,5]

[1]Centre for Public Health Research, Massey University, Wellington, New Zealand
[2]Occupational Medicine Specialists, Auckland, New Zealand
[3]Centre for Research in Environmental Epidemiology (CREAL), Barcelona, Spain
[4]School of Public Health, University of California, Berkeley, USA
[5]Department of Medical Statistics, London School of Hygiene and Tropical Medicine, London, UK

Correspondence to
Dr Andrea 't Mannetje, Centre for Public Health Research, Massey University, P.O. Box 756, Wellington 6140, New Zealand; a.mannetje@massey.ac.nz

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ABSTRACT

Objectives Exposure to 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) has inconsistently been associated with a decreased sex ratio of the offspring (number of male births divided by total births). We conducted a study among men and women who were employed in a New Zealand phenoxy herbicide production plant between 1969 and 1984, to study their offspring sex ratio in relation to their back-calculated TCDD serum concentrations determined in 2007/2008.

Methods A total of 127 men and 21 women reported that 355 children were conceived after starting employment at the plant. The association between their lipid-standardised TCDD serum concentrations back-calculated to the time of their offspring's birth and the probability of a male birth was estimated through logistic regression, adjusting for the age of the exposed parent at birth, current body mass index and smoking.

Results The overall sex ratio was 0.55 (197 boys, 158 girls). For fathers with serum TCDD concentrations ≥20 pg/g lipid at time of birth, the sex ratio was 0.47 (OR 0.49; 95% CI 0.30 to 0.79). The probability of a male birth decreased with higher paternal serum TCDD at time of birth (<4; 4–20; 20–100; ≥100 pg/g lipid), with ORs of 1.00 (reference); 1.00 (95% CI 0.50 to 2.02); 0.52 (95% CI 0.29 to 0.92); 0.45 (95% CI 0.23 to 0.89), p trend 0.007. For exposed mothers, the sex ratio was not reduced.

Conclusions This study indicates that paternal serum TCDD concentrations in excess of an estimated 20 pg/g lipid at time of conception are associated with a reduced sex ratio.

What this paper adds

- In 1996, it was first reported that fewer boys than girls were born from parents exposed to 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) in a 1976 industrial accident in Seveso, Italy. The reduction in sex ratio was present for exposed fathers and strongest when exposure occurred before the age of 19.
- Since then, sex ratios of only a handful of highly exposed populations have been reported, with inconsistent results.
- In this study of an occupationally exposed population, paternal TCDD serum levels in excess of 20 pg/g lipid at time of birth were associated with the birth of fewer boys than girls.
- This study indicates that TCDD's impact on the offspring sex ratio is dose-dependent and male-mediated, and suggests that the relevant time window of exposure extends into adulthood.

INTRODUCTION

Dioxins and, in particular, 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD), have been associated with a wide range of medium-term to long-term health effects.[1] In 1996, it was first reported that the sex ratio (male births divided by the total number of births) was reduced for parents exposed to TCDD in an industrial accident in 1976 in Seveso, Italy.[2] This association has since been examined in animal and human studies aiming to replicate this finding in other populations, find a mechanistic explanation, and determine the relevant dose and time window of exposure.

The number of studies in humans has been limited due to the small number of exposed cohorts for which individual TCDD serum measurements are available. In 1998, sex ratios for children fathered by US veterans of operation Ranch Hand, the unit responsible for spraying dioxin-contaminated agent orange in Vietnam from 1962 to 1971,[3] were reported. A lower sex ratio related to serum TCDD concentrations at time of conception was not observed.

In 2000, further findings of the Seveso study[4] indicated that a significantly lower sex ratio was present if fathers had been exposed to dioxin, particularly if they were younger than 19 years at the time of exposure (sex ratio 0.38). TCDD concentrations of mothers were not a significant predictor of the probability of a male birth. This suggested that the effect was male-mediated, and particularly strong if the initial exposure had occurred before or during puberty of the future fathers.

In response to this finding, the sex ratio of the offspring of the male Austrian chloracne cohort exposed to TCDD during production of 2,4,5-T in the early 1970s (n=157) was reported[5] with results consistent with the Seveso study, but based on small numbers.

In 2001, the sex ratio of offspring of 281 male workers from two US plants producing phenoxy herbicides (including 2,4,5-T) was reported.[6] Adult

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TCDD exposure at time of conception was not associated with a reduced sex ratio.

Finally, in 2002, a report on Russian phenoxy herbicide producers from Ufa was published,[7] indicating a decrease in the number of boys for exposed fathers, while no decrease was observed for exposed mothers, in line with the Seveso findings, although fathers were older than 20 at the time of exposure.

In summary, offspring sex ratios have now been reported for four sizeable populations with known exposure to TCDD. For two (US-Ranch Hand, US-National Institute for Occupational Safety and Health (NIOSH)), a null association was reported and for two (Italy-Seveso, Russia-Ufa), a lower sex ratio was reported to be associated with exposure in the fathers but not for the mothers.

Here we report on the sex ratio of the offspring of male and female workers employed at a phenoxy herbicide production plant in New Plymouth, New Zealand between 1969 and 1984,[8] for whom individual TCDD serum concentration were determined in 2007/2008 (serum concentrations of TCDD of this population have been reported previously[9]).

METHODS

The workers of a phenoxy herbicide production plant included in this study were part of the New Zealand component of the International Agency for Research on Cancer (IARC) international cohort of producers and sprayers of phenoxy herbicides,[8] and had worked for at least 1 month between 1969 and 1984 in the pesticide production plant in New Plymouth, New Zealand. Of the 1025 original production cohort members, 631 were known to be alive, living in New Zealand and aged <80 years on 1 January 2006. Of these, 430 were randomly selected and invited to participate in a morbidity survey, and 244 completed the survey. During 2007/2008, the participants provided blood for the determination of TCDD serum concentrations of which detailed results have been reported elsewhere.[9] During a face-to-face interview, participants were asked to provide details on all of their live-born or stillborn biological children, including sex, name, date and place of birth. The questionnaire did not ask about specific health outcomes, but a free-text comments box was available for comments regarding the health of the child. Based on the responses, dichotomous variables of the offspring's-specific health outcomes at any time after birth were constructed for those reported by multiple study participants.

Serum samples taken at time of interview were analysed for concentrations of TCDD, 6 other chlorinated dibenzodioxins, 10 chlorinated dibenzofurans and 15 polychlorinated biphenyls (PCBs), using gas chromatography-high-resolution mass spectrometry. Individual serum concentrations of TCDD at the time of phlebotomy were back-calculated to the time of the birth of the child by using a first-order elimination model[9] and a half-life of 7.6 years, which was determined based on the Ranch Hand Veteran population,[10] of which TCDD serum levels were found to be similar to the population studied here.[9] Overall, the cohort had a mean serum TCDD concentration of 9 pg/g, which back-calculated to 1987, the last year of 2,4,5-T production in this plant, would have been 49 pg/g. Highest TCDD serum concentrations were found for the group of 60 producers directly involved in phenoxy herbicide production (mean 19 pg/g which back-calculated to 1987 would have been 109 pg/g), while most other people working within the plant had serum concentrations of dioxin-like compounds comparable to those of the general population.[9]

Exposure variables were based on (1) serum TCDD concentration at time of phlebotomy and (2) back-calculated serum TCDD concentration to the time of birth of the offspring.

Sex ratios for exposure groups were calculated as the number of male births divided by the total number of births. The association between exposure and the probability of a male birth was estimated through logistic regression of correlated outcome data with children from the same parent being correlated (using SAS proc genmod with repeated statement (Base SAS V.9.3 Procedures Guide Cary. NC, USA: SAS Institute Inc., 2011)), crude and adjusting for the age of exposed parent at year of birth, current body mass index (BMI) and smoking status of the parent (as they can be associated with TCDD serum levels and, in the case of age and BMI of the parent, also with offspring sex ratio). The association between the natural logarithm of the estimated serum TCDD concentration at birth (ln-TCDD) and the probability of a parent-reported health problem in the offspring was estimated through logistic regression of correlated outcome data, adjusting for sex of the exposed parent, age of exposed parent at time of birth and sex of the child.

This study received ethical approval of the Central Regional Ethics Committee (ref: CEN/06/02/002) on 19 May 2006. All study participants provided informed consent.

RESULTS

Of the 244 participants, 32 did not report having biological children. A total of 212 participants reported a total of 622 births (175 fathers with 509 births, 2.9 births on average, 37 mothers with 113 births, 3.1 births on average). For 10 of the 622 births, sex was not reported and was excluded from the analyses. A further 257 were excluded from the analyses because they were conceived before the parent started employment at the plant, leaving 355 births from 127 fathers and 21 mothers for the analyses. The overall sex ratio was 0.55 for children of fathers employed at the plant and 0.59 for mothers (table 1). None of the children had both parents employed at the plant.

Table 2 presents the results of the logistic regressions for correlated data, modelling the probability of a male birth and stratified by sex of the parent employed at the plant.

Having a serum TCDD concentration ≥4 pg/g lipid at the time of phlebotomy was associated with a decreased probability of a male birth for exposed fathers (OR 0.46, 95% CI 0.29 to 0.73), with indication of a dose–response relationship (p trend=0.01). When using categories of serum TCDD concentration back-calculated to the time of birth, a stronger dose–response association was observed (p trend=0.007). Compared with the reference group of fathers with back-calculated TCDD levels below 4 pg/g at time of birth, fathers with serum TCDD of 4–20 pg/g did not have lower offspring sex ratio, while a serum TCDD of 20–100 pg/g at birth was associated with an OR of 0.52 (95% CI 0.29 to 0.92) and ≥100 pg/g with an OR of 0.45 (95% CI 0.23 to 0.89). For exposed mothers, the opposite pattern to that of exposed fathers was observed (ie, higher probability of a male birth with higher maternal TCDD), but ORs were not statistically significant and based on a small number of births.

Table 3 presents the association between the father's serum TCDD concentration back-calculated to the time of birth and the probability of a male birth, stratified by the age of first paternal exposure, assuming exposure started at the start of employment at the plant. These results suggest a stronger association between TCDD exposure and the probability of a male birth if first exposure occurred before the age of 37.

Table 1 Study population characteristics: number of births by plant employees conceived after starting employment, by parents' demographic characteristics

	Fathers employed at plant (n=127)			Mothers employed at plant (n=21)		
	Girls (n)	Boys (n)	Sex ratio	Girls (n)	Boys (n)	Sex ratio
	137	167	0.55	21	30	0.59
Year of birth						
<1970	21	14	0.40			
70–80	54	61	0.53	5	14	0.74
80–90	42	61	0.59	13	12	0.48
>1990	20	31	0.61	3	4	0.57
Age of parent (employed at plant) at birth						
<25	19	20	0.51	3	11	0.79
25–30	47	56	0.54	7	6	0.46
30–40	59	80	0.58	9	11	0.55
≥40	12	11	0.48	2	2	0.50
Age of other parent at birth						
<25	33	44	0.57			
25–30	47	51	0.52			
30–40	30	43	0.59			
≥40	3	1	0.25			
Unknown	24	28	0.54	21	30	0.59
Age at start of employment						
23–30	26	39	0.60	7	6	0.46
30–37	47	66	0.58	8	11	0.58
>37	64	62	0.49	6	13	0.68

Table 2 Association between TCDD exposure of the parent and male birth outcome

	Fathers employed at plant (n=127)							Mothers employed at plant (n=21)				
	Girls (n)	Boys (n)	Sex ratio	Crude OR	95% CI	OR*	95% CI	Girls (n)	Boys (n)	Sex ratio	Crude OR	95% CI
Serum TCDD of parent at time of phlebotomy												
<4 pg/g lipid	76	114	0.60	1.00	ref	1.00	ref	14	18	0.56	1.00	ref
≥4 pg/g lipid	61	53	0.46	0.58	0.37 to 0.91	0.46	0.29 to 0.73	7	12	0.63	1.40	0.66 to 2.97
<2 pg/g lipid	54	80	0.60	1.00	ref	1.00	ref	6	7	0.54		
2–4 pg/g lipid	22	34	0.61	1.06	0.57 to 1.97	1.10	0.59 to 2.05	8	11	0.58		
4–8 pg/g lipid	39	28	0.42	0.48	0.28 to 0.84	0.44	0.26 to 0.75	7	10	0.59		
≥8 pg/g lipid	22	25	0.53	0.79	0.42 to 1.46	0.56	0.28 to 1.10	0	2	–		
				p trend	0.083	p trend	0.010					
Estimated serum TCDD of parent at time of birth												
<20 pg/g lipid	74	112	0.60	1.00	ref	1.00	ref	15	17	0.53	1.00	ref
≥20 pg/g lipid	63	55	0.47	0.58	0.37 to 0.90	0.49	0.30 to 0.79	6	13	0.68	1.90	0.84 to 4.31
<4 pg/g lipid	59	88	0.60	1.00	ref	1.00	ref	9	9	0.50		
4–20 pg/g lipid	15	24	0.62	1.08	0.53 to 2.18	1.00	0.50 to 2.02	6	8	0.57		
20–100 pg/g lipid	30	27	0.47	0.60	0.35 to 1.02	0.52	0.29 to 0.92	6	12	0.67		
≥100 pg/g lipid	33	28	0.46	0.58	0.30 to 1.09	0.45	0.23 to 0.89	0	1	–		
				p trend	0.037	p trend	0.007					

*OR (modelling the probability of a male birth) adjusted for: age of parent at year of birth, current BMI of parent, smoking status of parent.
BMI, body mass index; TCDD, 2,3,7,8-tetrachlorodibenzo-p-dioxin.

We investigated which of the variables included as confounders resulted in the largest change from the crude ORs for the association between TCDD exposure categories at the time of birth and the probability of a male birth for exposed fathers (see online supplementary table S1). The only variable resulting in an appreciable change of the crude OR was current BMI of the father, which itself was associated with a higher probability of a male birth. Inclusion of smoking of the parent employed at the plant, age of mother at birth, age of father at birth and year of birth of the child did not appreciably change the results.

Additional stratifications were performed for the association between the father's serum TCDD concentration back-calculated to the time of birth and the probability of a male birth, to evaluate whether the association between TCDD and

Table 3 Association between estimated paternal serum TCDD concentration at time of birth and the probability of a male offspring, stratified by father's age of first exposure (estimated by father's age at start of employment)

Fathers employed at plant (n=127)							
Age of father at start of employment	**Girls (n)**	**Boys (n)**	**Sex ratio**	**Crude OR**	**95% CI**	**OR***	**95% CI**
23–30							
<4 pg/g lipid	15	24	0.62	1.00	ref	1.00	ref
4–20 pg/g lipid	4	9	0.69	1.37	0.36 to 5.31	2.00	0.35 to 11.4
20–100 pg/g lipid	4	5	0.56	0.51	0.16 to 1.68	0.17	0.04 to 0.68
≥100 pg/g lipid	3	1	0.25	0.20	0.03 to 1.37	0.04	0.01 to 0.26
				p trend	0.128	p trend	0.016
30–37							
<4 pg/g lipid	24	43	0.64	1.00	ref	1.00	ref
4–20 pg/g lipid	3	5	0.63	0.94	0.19 to 4.75	1.26	0.20 to 8.08
20–100 pg/g lipid	12	11	0.48	0.51	0.25 to 1.07	0.37	0.16 to 0.85
≥100 pg/g lipid	8	7	0.47	0.49	0.12 to 2.09	0.59	0.18 to 1.95
				p trend	0.148	p trend	0.062
>37							
<4 pg/g lipid	20	21	0.51	1.00	ref	1.00	ref
4–20 pg/g lipid	8	10	0.56	1.20	0.42 to 3.48	2.02	0.78 to 5.21
20–100 pg/g lipid	14	11	0.44	0.75	0.28 to 2.05	0.87	0.34 to 2.25
≥100 pg/g lipid	22	20	0.48	0.86	0.35 to 2.12	1.22	0.48 to 3.13
				p trend	0.625	p trend	0.934

*OR (modelling the probability of a male birth) adjusted for: age of parent at year of birth, current BMI of parent, smoking status of parent.
BMI, body mass index; TCDD, 2,3,7,8-tetrachlorodibenzo-p-dioxin.

sex ratio was observed independently of the following variables (see online supplementary tables S2–S4): (1) father's age at birth; (2) father's current BMI; (3) year of birth of the child. These analyses indicated that the negative association between TCDD and the probability of a male birth was observed independently of the age of the father, while the association was strongest for fathers with a BMI<25 (p trend 0.02) and for births after 1980 (p trend <0.001).

Of the 355 births that occurred after the start of employment of the parent at the plant, a health problem was reported for 57 births (table 4). Specific health problems that were repeatedly reported included asthma, birth defects and thyroid/gland problems. All health problems combined (any health problem reported) was not significantly associated with elevated serum TCDD, nor was it significantly associated with ln-TCDD. Congenital malformations in the offspring were not more frequently reported by highly exposed parents (OR 0.54; 95% CI 0.16 to 1.85) and were not associated with ln-TCDD (OR 0.89, 95% CI 0.68 to 1.16). Asthma was reported for nine children, but an OR could not be calculated due to the small numbers. None of the parents exposed to <20 pg/g TCDD at time of birth reported a thyroid problem in offspring born after the start of employment, while of the 137 children born to highly exposed parents, a thyroid problem was reported for three (three children of two fathers and one mother with serum TCDD at time of birth of 79, 90 and 208 pg/g, respectively). Although based on very small numbers, the reporting of a thyroid problem in the offspring was positively associated with ln-TCDD at time of birth (OR 1.85, 95% CI 1.37 to 2.48).

DISCUSSION

In this study, serum TCDD concentrations above 20 pg/g lipid at the time of birth for occupationally exposed fathers were associated with the birth of relatively fewer boys than girls. For occupationally exposed mothers, the association was in the opposite direction from males, and while not statistically significant and based on much smaller numbers, this would be in line with the hypothesis that hazards affecting endocrine hormones may have opposite effects on sex ratio in men and women.[11] Overall, these findings support a male-mediated reduction in sex

Table 4 Association between an estimated parental ≥20 pg/g TCDD serum concentration at time of birth and the probability of a parent-reported health problem in the offspring

355 births after start of employment						
	TCDD<20 pg/g (n=218)		**TCDD≥20 pg/g (n=137)**		**OR* (95% CI)**	**OR for ln-TCDD continuous (95% CI)**
Any health problem reported	33	15%	24	18%	1.33 (0.72 to 2.45)	1.10 (0.91 to 1.32)
Congenital malformation	13	6%	5	4%	0.54 (0.16 to 1.85)	0.89 (0.68 to 1.16)
Thyroid/gland problem†	0	0%	3	2%	–	1.85 (1.37 to 2.48)

*OR adjusted for: age of parent at year of birth, sex of exposed parent, sex of child.
†Thyroid/gland problem refers to the parent reporting that their offspring had 'gland problems', 'thyroid problems' or 'thyroid surgery'. For none of the offspring with reported thyroid/gland problem, the parent reported they had a thyroid problem themselves. Of the three children reported to have thyroid problems, two were from an exposed father and one from an exposed mother. The OR could not be adjusted for age of exposed parent at year of birth.
TCDD, 2,3,7,8-tetrachlorodibenzo-p-dioxin.

ratio associated with serum TCDD concentration at the time of birth, consistent with the findings of the Seveso study[4] and the Russian pesticide producers study.[7]

The sex ratio of offspring was 0.47 for fathers with serum TCDD concentrations above 20 pg/g at the time of birth. This is comparable to, but not as low as, the sex ratios reported for Seveso or the Russian pesticide producers (table 5), for whom TCDD exposures were generally higher than in the New Zealand producers.[9]

Although this population was occupationally exposed to a variety of compounds and pesticides (besides phenoxy herbicides also, eg, triazines, surfactants, solvents), we consider paternal TCDD exposure at the time of conception to be the most likely explanation for the observed association, given the consistency of our findings with those reported for the Seveso population (where TCDD was the only exposure measured), and the presence of a dose–response relationship for TCDD at time of birth.

The mechanism through which paternal TCDD exposure could affect the sex ratio has not yet been established, but recent animal and human studies provide some insight. In a study of TCDD-treated male mice mated with non-treated females,[12] the Y-bearing/X-bearing sperm ratio was not significantly decreased, but the sex ratio of the two-cell embryos of the TCDD group was significantly lower than that of the control group. In a study of male rats which had in utero TCDD exposure and were mated with unexposed females, the number of male offspring was significantly decreased.[13] Another study in rats[14] did not observe a change in sex ratio, but no distinction between paternal and maternal TCDD exposure was made. Thus, while data are limited, some studies in rodents support the hypothesis of a male-mediated reduction in sex ratio resulting from TCDD exposure starting either in utero or at reproductive age. The effect is not explained by changes in the Y-bearing/X-bearing sperm ratio,[13] nor by a disproportionate loss of male embryos, and it has been proposed that a decrease in fertility of Y-bearing sperm may be responsible.[13]

Studies of the effects of TCDD on semen in exposed human populations are limited. In the Seveso study, in utero and lactational TCDD exposure of children of exposed mothers was associated with a permanent reduction in sperm quality (lower sperm concentration, count and motility).[15] This effect was also observed in rhesus monkeys.[16] Seveso men exposed to TCDD in infancy also had reduced sperm concentration and motility,[17] while the opposite effect was seen with exposure during puberty. No effect on semen quality was seen in men exposed to TCDD as adults.[17] In a study of veterans of operation Ranch Hand, no associations between serum dioxin levels and testicular abnormalities, sperm count, sperm abnormalities or testicular volume were observed.[18] Thus, while there is evidence of negative effects on sperm quality when TCDD exposure occurs at young ages (in utero to puberty), there are currently no studies indicating that adult exposure to TCDD affects sperm quality.

The relevant time window of the father's exposure's impact on the offspring's sex ratio has also been investigated. The Seveso study found that reduction in sex ratio was strongest when exposure occurred before the age of 19.[4] In occupationally exposed cohorts such as this study and the Russian study, timing of first exposure is difficult to establish, but it is safe to assume that first employment and therefore first exposure would have occurred in adulthood (the youngest age of first employment at the plant for this study population was 23). Thus, our findings, as well as those of the Russian study,[7] indicate that the effect is not limited to exposure before or during puberty, although we did observe the strongest association for fathers first exposed before the age of 37.

The absence of TCDD serum measurements at the time of birth is a limitation of this study. We had to rely on serum TCDD concentrations determined in 2007/2008 back-calculated to the time of birth, which could be more than 30 years prior to phlebotomy. We had to assume that exposure started when employment started, which may not be the case for all participants, potentially resulting in a substantial overestimation of TCDD exposure for those births that occurred closest after the start of employment. Since exposure misclassification could obscure associations, this may explain the absence of a dose–response association for the offspring born prior to 1980, while a clear dose–response was observed for offspring born after 1980, as for the latter exposure misclassification is likely to be less substantial. In our back-calculation, we also assumed that elimination rates were the same for all, although higher body percentage fat has been associated with slower elimination of TCDD,[10] resulting in a relative overestimation of historical TCDD exposure in those with high BMI. This may possibly explain the weak dose–response association for overweight fathers, while we did observe a strong dose–response association for fathers who were not overweight.

We considered a range of factors that may confound the association between TCDD and sex ratio, of which only paternal BMI was found to alter the TCDD–sex ratio association after inclusion in the model. Paternal BMI was positively associated with a male birth outcome: sex ratio of fathers with normal weight (BMI<25) was 0.47, while for overweight fathers (BMI≥25) it was 0.60, a pattern which has been observed previously.[19] Inclusion of current paternal BMI in the model strengthened the negative association between paternal TCDD and male birth outcome, possibly due to BMI-associated misclassification of exposure. Adjustment for BMI could, however, not explain the observed associations, given that the crude models indicated the same trends.

In this population, the overall sex ratio was higher (0.55) than expected for the general population (generally 0.51) with

Table 5 Sex ratios reported for the offspring of TCDD exposed populations

	Comparison group		TCDD exposed group		
	Children (n)	Sex ratio	Paternal TCDD at the time of conception of the TCDD exposed group	Children (n)	Sex ratio
US, Ranch Hand[3]	346	0.51	≥10 pg/g	557	0.51
Italy, Seveso[4]	271	0.56	>15 pg/g	403	0.44
US, NIOSH[6]	292	0.51	≥20 pg/g	252	0.56
Russia, Ufa[7]	Ufa city	0.51	Not reported	188	0.38
New Zealand (this study)	186	0.60	≥20 pg/g	118	0.47

NIOSH, National Institute for Occupational Safety and Health; TCDD, 2,3,7,8-tetrachlorodibenzo-p-dioxin.

higher sex ratios for recent years of birth, for which the reasons are not clear. Although sex ratios have slightly increased over time in New Zealand,[20] this cannot explain the magnitude observed here. Also, the effect is contrary to a parental age cohort effect that could be expected: the earlier births in this cohort are more likely to be from younger parents and have lower birth order, which have both been associated with a higher sex ratio.[21] A paternal BMI-related cohort effect is consistent with the observed increase in sex ratio over time, given that higher paternal BMI has been associated with a higher sex ratio.[19] Although it is probable that an increase in the fathers' BMI over time could have occurred, we do not have data to confirm this. Alternatively, the observed increase in sex ratio over time could in part reflect a real effect of TCDD exposure, with the highest TCDD exposure and lowest sex ratio to be expected in the earlier years, as also seen in the Seveso study.[5]

In this study, we did not observe that a higher serum TCDD concentration was associated with a more frequent report of congenital malformations in the offspring. Most evidence of health effects in offspring is based on studies involving in utero or perinatal TCDD exposure, with the mother as the main route of exposure.[1] In this study, TCDD serum concentrations were available mainly for fathers (we did not determine the mother's TCDD serum concentration of male cohort members) and the number of female cohort members was insufficient to study the association between maternal TCDD exposure and reported health outcomes in the offspring. In addition, the study used the parent's self-report of health problems in the offspring based on an open-ended question, which is likely to be subject to substantial misclassification and lacks clinical verification. Notwithstanding these limitations, we did observe an association between continuous TCDD (ln-TCDD at time of birth) and reported thyroid problems in the offspring. Although this association is based on very small numbers, it is statistically significant and is noteworthy in the light of toxicological and mechanistic data indicating that dioxin may impair thyroid function in the offspring.[22] Evidence in human populations is very limited, but it has been reported that children born from mothers exposed to TCDD in the Seveso incident had higher neonatal blood thyroid-stimulating hormone (b-TSH, a sensitive marker of subclinical primary hypothyroidism) than the reference population, and maternal TCDD levels estimated at the date of delivery were positively associated with neonatal b-TSH.[23] After further testing, two children from the contaminated areas and none from the reference were diagnosed with primary hypothyroidism.[23] Thus, thyroid effects in the offspring associated with parental TCDD exposure may be of clinical significance and warrants further investigation.

CONCLUSIONS

This study lends further support to a second-generation effect of TCDD exposure, with paternal TCDD serum concentrations in excess of an estimated 20 pg/g lipid associated with the birth of relatively fewer boys than girls.

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Sex ratio of the offspring of New Zealand phenoxy herbicide producers exposed to 2,3,7,8-tetrachlorodibenzo-p-dioxin

Andrea 't Mannetje, Amanda Eng, Chris Walls, Evan Dryson, Manolis Kogevinas, Collin Brooks, Dave McLean, Soo Cheng, Allan H Smith and Neil Pearce

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Notes