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Associations of dioxins, furans and dioxin-like PCBs with diabetes and pre-diabetes: Is the toxic equivalency approach useful? ☆

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ABSTRACT

Toxic equivalency factors for dioxins and dioxin-like compounds have been established by the World Health Organization. Toxic equivalency (TEQ) was derived using 6 chlorinated dibenzo-*p*-dioxins, 9 chlorinated dibenzofurans and 8 polychlorinated biphenyls, in blood, from the 1999–2004 National Health and Nutrition Examination Survey. Relationships of 8 individual chemicals, the number of compounds elevated, and TEQ with pre-diabetes and total diabetes (diagnosed and undiagnosed) were investigated using logistic regressions. For the 8 chemicals analyzed separately, values above the 75th percentile were considered elevated, whereas for the other 15 compounds, values above the maximum limit of detection were considered elevated. Pre-diabetes with glycohemoglobin (A1c) 5.9–6.4% was associated with PCB 126, PCB 118 and having one or more compounds elevated (odds ratio 2.47, 95% CI 1.51–4.06). Pre-diabetes with A1c 5.7–5.8% was not associated with any individual chemical or the number of compounds elevated. Total diabetes was associated with 6 of the 8 individual compounds tested, and was associated with having 4 or more compounds elevated. Toxic equivalency $\geq$81.58 TEQ fg/g was associated with total diabetes (odds ratio 3.08, 95% CI 1.20–7.90), but was not associated with A1c 5.9–6.4%. Having multiple compounds elevated appeared to be important for total diabetes, whereas for pre-diabetes with A1c 5.9–6.4%, having a single compound elevated appeared most important. Diabetes plus A1c $\geq$5.9% was associated with 34.16–81.57 TEQ fg/g (odds ratio 2.00, 95% CI 1.06–3.77) and with $\geq$81.58 TEQ fg/g (odds ratio 2.48, 95% CI 1.21–5.11), indicating that half the population has elevated risk for this combination of conditions.

1. Introduction

The World Health Organization has established toxic equivalency factors (TEF) for 29 dioxins and dioxin-like compounds (Van den Berg et al., 2006). These TEFs compare the toxicity of a chemical to the toxicity of 2,3,7,8-tetrachlorodibenzo-*p*-dioxin. Toxic equivalency (TEQ) of the 29 chlorinated dibenzo-*p*-dioxins, chlorinated dibenzofurans, and dioxin-like polychlorinated biphenyls (PCB) is computed by multiplying the concentration of each chemical with its TEF, and summing the products. The present TEF scheme and the TEQ methodology are primarily intended for estimating exposure and risks via oral ingestion (Van den Berg et al., 2006).

States related to glucose homeostasis include metabolic syndrome, insulin resistance, pre-diabetes, and diabetes. In the environmental epidemiology literature on dioxin and dioxin-like compounds, much attention has been given to diabetes, metabolic syndrome, and insulin resistance, but less to pre-diabetes (Frithsen and Everett, 2012). The most up-to-date definition of pre-diabetes involves the measurement of blood glycohemoglobin (A1c), with a value in the range 5.7–6.4% indicating pre-diabetes. A1c $\geq$6.5% is also used to define undiagnosed diabetes (ADA, 2010).

A number of cross-sectional studies on dioxin-like compounds and diabetes have been conducted. A study in a First Nation Community in Northern Ontario, Canada evaluated several PCBs and *p,p′*-DDE (*p,p′*-dichlorodiphenyltrichloroethylene) and their relationship with diabetes. Both wet weight and lipid-standardized PCBs were tested and found to be associated with diabetes (Philibert et al., 2009). Another study examined 111 chemicals, including PCBs and *p,p′*-DDE, among Great Lakes sport fish consumers who were surveyed and had blood collected in 2004–2005. The sum of dioxin-like PCBs was associated with A1c >6.3% in some adjusted analyses, but not when further adjusted for *p,p′*-DDE (Turyk et al., 2009a, b). In a Slovakian study of persons living in a heavily polluted area, PCBs were associated with pre-diabetes (impaired fasting glucose and/or impaired

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glucose tolerance) and diabetes. Stepwise logistic regressions showed that PCBs were a significant factor for pre-diabetes (Ukropec et al., 2010). In Japan, a population-based study expressed 29 dioxins, furans, and dioxin-like PCBs as TEQs. The highest quartile of total TEQ had an elevated odds ratio for diagnosed plus undiagnosed diabetes in an adjusted logistic regression (Uemura et al., 2008).

Several pertinent cross-sectional analyses have been done out of the US National Health and Nutrition Examination Study (NHANES). Of special note, Patel et al. (2010) studied associations of 266 environmental contaminants with diabetes in the NHANES 1999–2006. Evaluating each 2-year cohort separately, they found 2 dioxins, one furan and one dioxin-like PCB to be associated with diabetes in one of the three cohorts. However, Patel et al. (2010) considered environmental factors significant only when they were associated with diabetes in two cohorts. In an earlier study, Lee et al. (2007) evaluated the relationship of 3 dioxins, 3 furans and 4 dioxin-like PCBs, with diabetes, in the NHANES 1999–2002. They included PCB 74 as a dioxin-like PCB, even though it is not considered dioxin-like by Van den Berg et al. (2006). Lee et al. (2007) also summed each type of chemical (dioxin, furan, and dioxin-like PCB), but did not combine them as is done with the TEQ methodology. None of the NHANES studies look at the relationship of dioxin and dioxin-like compounds, and pre-diabetes, though Everett and Matheson (2010) found 2 pesticides, *p,p′*-DDT (*p,p′*-dichlorodiphenyltrichloroethane) and heptachlor epoxide, associated with pre-diabetes in the NHANES 1999–2004.

A few longitudinal studies of dioxin and dioxin-like compounds, and diabetes, have been conducted. A case-control study assessed the effects of spraying Agent Orange and 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD)-contaminated herbicides in Vietnam. Veterans exposed to more TCDD than the background concentration, whose service was in 1969 or earlier, and/or consisted of $\geq$90 days of spraying, were more likely to develop diabetes than the comparison group (Michalek and Pavuk, 2008). A Taiwanese study with 24 years of follow-up evaluated those persons who were accidentally exposed to high levels of PCBs and furans by ingestion of contaminated rice-bran oil. In age-adjusted logistic regressions, women in the exposed group but not men, had an increased risk of developing diabetes (Wang et al., 2008). An additional cohort study of Great Lakes sport fish consumers followed participants from 1994–1995 to 2004–2005. The sum of the PCBs and PCB 118 were not associated with incident diabetes in adjusted analyses (Turyk et al., 2009a,b). Data from the Coronary Artery Risk Development in Young Adults (CARDIA) cohort was used to examine the association of 22 PCB congeners and nine other persistent organic pollutants, and incident diabetes. Dioxin-like PCBs 105, 118, 156, 157 and 167 were not associated with incident diabetes (Lee et al., 2010).

The purpose of our study was to test the World Health Organization's TEQ methodology in the NHANES 1999–2004, and to investigate relationships with pre-diabetes. While Van den Berg et al. (2006) identifies 29 dioxin and dioxin-like compounds, we were limited to 23 which were measured in the NHANES 1999–2000. While it is desirable to evaluate pesticides along with dioxins, furans and dioxin-like PCBs, this is not possible using the NHANES 2003–2004 data. Pesticides were measured in "Subsample B," while dioxins, furans and PCBs were measured in "Subsample C."

2. Materials and methods

We used data derived from the National Health and Nutrition Examination Survey (NHANES), 1999–2004, for this study. Since 1999, and continuing to the present, the NHANES has visited 15 communities per year across the United States. A nationally representative sample of the noninstitutionalized US population, the NHANES design includes an over-sampling of minorities and an ability to make population estimates. More information on the methodology of the NHANES 1999–2004, including laboratory assessment, can be found at the National Center for Health Statistics (NCHS) website (CDC, 2012).

We investigated the association of dioxins and dioxin-like compounds with pre-diabetes, and total diabetes (diagnosed plus undiagnosed). Pre-diabetes was defined as persons who had glycohemoglobin (A1c) 5.7–6.4% (ADA, 2010). We divided the persons who had pre-diabetes into two groups of approximately equal size, with "low" pre-diabetes defined as A1c 5.7–5.8%, and "high" pre-diabetes defined as A1c 5.9–6.4%. Diagnosed diabetes was determined by self-report answer to the question: "Other than during pregnancy, have you ever been told by a doctor or health professional that you have diabetes or sugar diabetes?" Those who answered "borderline" were considered to not have diabetes. Undiagnosed diabetes was defined as persons who had A1c $\geq$6.5% who had not been diagnosed as having diabetes (ADA, 2010). We did not use fasting plasma glucose for determination of undiagnosed diabetes because glucose was measured on a fasting subsample of participants and would have reduced by half the number of persons in our analyses.

Six chlorinated dibenzo-*p*-dioxins, 9 chlorinated dibenzofurans, and 8 dioxin-like polychlorinated biphenyls (Table 1) were measured in nonfasting blood samples of a one third, stratified random, subsample of participants 12 years old and older. We evaluated persons in this subsample who were $\geq$20 years old. The unweighed number of participants in our study were 2611 which represented a total of 118,631,242 Americans (Table 2). The 23 dioxins and dioxin-like compounds were measured in serum by high-resolution gas chromatography/isotope-dilution high-resolution mass spectrometry. Concentrations were expressed per gram of blood, rather than adjusting for lipids, as recommended by Schisterman et al. (2005). Each person had a sample-specific limit of detection. Values below or equal to, the maximum limit of detection observed (Table 1) were set to zero for the purposes of our analyses. Eight of the 23 dioxins and dioxin-like compounds had >25% of their values above the maximum limit of detection, and were analyzed separately. The 8 dioxins and dioxin-like chemicals were categorized as less than or equal to the maximum limit of detection, greater than or equal to the 75th percentile, and an intermediate category, greater than the maximum limit of detection and less than the 75th percentile. We also evaluated the number of compounds elevated. A compound was considered elevated if its concentration was above the 75th percentile for the 8 chemicals listed in Tables 3 and 4, or above the maximum limit of detection for the other 15 compounds. Finally, total TEQ was calculated by multiplying concentrations and toxic equivalency factors

Table 1
Dioxins and dioxin-like compounds investigated.

	Maximum limit of detection
Dioxins	
2,3,7,8-Tetrachlorodibenzo-*p*-dioxin (TCDD)	47.73 fg/g
1,2,3,7,8-Pentachlorodibenzo-*p*-dioxin (PeCDD)	53.81 fg/g
1,2,3,6,7,8-Hexachlorodibenzo-*p*-dioxin (HxCDD)	88.60 fg/g
1,2,3,7,8,9- Hexachlorodibenzo-*p*-dioxin (HxCDD)	81.46 fg/g
1,2,3,4,6,7,8-Heptachlorodibenzo-*p*-dioxin (HpCDD)	157.33 fg/g
1,2,3,4,6,7,8,9-Octachlorodibenzo-*p*-dioxin (OCDD)	1945.82 fg/g
Furans	
2,3,7,8-Tetrachlorodibenzofuran (TCDF)	51.90 fg/g
1,2,3,7,8-Pentachlorodibenzofuran (PeCDF)	47.87 fg/g
2,3,4,7,8-Pentachlorodibenzofuran (PeCDF)	51.12 fg/g
1,2,3,4,7,8-Hexachlorodibenzofuran (HxCDF)	47.31 fg/g
1,2,3,6,7,8-Hexachlorodibenzofuran (HxCDF)	49.29 fg/g
1,2,3,7,8,9-Hexachlorodibenzofuran (HxCDF)	47.16 fg/g
2,3,4,6,7,8-Hexachlorodibenzofuran (HxCDF)	55.23 fg/g
1,2,3,4,6,7,8-Heptachlorodibenzofuran (HpCDF)	71.63 fg/g
1,2,3,4,6,7,8,9-Octachlorodibenzofuran (OCDF)	165.11 fg/g
PCBs	
3,4,4′,5-Tetrachlorobiphenyl (PCB 81)	274.85 fg/g
3,3′,4,4′,5-Pentachlorobiphenyl (PCB 126)	86.48 fg/g
3,3′,4,4′,5,5′-Hexachlorobiphenyl (PCB 169)	114.98 fg/g
2,3,3′,4,4′-Pentachlorobiphenyl (PCB 105)	0.048 ng/g
2,3′,4,4′,5-Pentachlorobiphenyl (PCB 118)	0.041 ng/g
2,3,3′,4,4′,5-Hexachlorobiphenyl (PCB 156)	0.048 ng/g
2,3,3′,4,4′,5′-Hexachlorobiphenyl (PCB 157)	0.048 ng/g
2,3′,4,4′,5,5′-Hexachlorobiphenyl (PCB 167)	0.048 ng/g

Table 2
Diabetes categories, number of compounds elevated, and toxic equivalency (TEQ) quartiles.

	Unweighed *N*	Population estimate	Proportion (%)
Diabetes category			
Normal	1961	96,505,171	81.3
Pre-diabetes	366	13,260,128	11.2
A1c 5.7-5.8%	192	6,949,135	5.8
A1c 5.9-6.4%	174	6,310,992	5.3
Total diabetes	284	8,865,944	7.5
Undiagnosed diabetes	54	1,799,295	1.5
Diagnosed diabetes	230	7,066,648	6.0
Number of Compounds Elevated[a]			
0	952	47,915,009	40.4
1	339	16,975,722	14.3
2	237	11,168,598	9.4
3	166	7,408,311	6.2
4–8	523	20,698,645	17.4
9–13	320	11,893,334	10.0
≥14	74	2,571,622	2.2
Toxic Equivalency (TEQ fg/g)			
<13.82	597	29,656,016	25.0
13.82–34.15	588	29,644,027	25.0
34.16–81.57	623	29,655,962	25.0
≥81.58	803	29,675,237	25.0
Total sample	2611	118,631,242	100

[a] Note that only 11.9% of the sample (unweighed $N=286$) had no compounds above the maximum limit of detection. The number of compounds elevated does not include values above the maximum limit of detection, and below the 75th percentile, for the 8 chemicals shown in Tables 3 and 4. For the other 15 chemicals, values above the maximum limits of detection are included.

established by the World Health Organization (Van den Berg et al., 2006). TEQ was classified using quartiles for logistic regression analyses.

We tested the associations of 8 individual chemicals, the number of compounds elevated, and TEQ, with "low" pre-diabetes (A1c 5.7–5.8%), "high" pre-diabetes (A1c 5.9–6.4%), total diabetes (diagnosed and undiagnosed), and diabetes plus A1c ≥5.9% in adjusted logistic regressions. These logistic regressions were adjusted for age, sex, race/ethnicity, education, poverty income ratio, body mass index, waist circumference, energy adjusted fruit and vegetable consumption, physical activity and family history of diabetes. Race/ethnicity was classified as Non-Hispanic White, Non-Hispanic Black, Mexican American, and other Hispanic. Education was classified as less than 9th grade, 9–12th grade without a diploma, high school graduate/GED or equivalent, some college or associate degree, and college graduate or above. Poverty income ratio (PIR) was analyzed as a continuous variable, and was the ratio of a family's income to their appropriate poverty threshold based on family size (US Census Bureau, 2012a). PIR was top coded at 5, and values below 1.00 were below the official poverty threshold (US Census Bureau, 2012b). Body mass index (kg/m^2) was derived from measured height and weight measurements collected in the NHANES physical examination. Waist circumference (cm) was also measured in the NHANES physical examination. Number of fruit and vegetable servings consumed per day was determined from questions in the NHANES dietary interview. This figure was then adjusted based on an estimate of energy intake (kcal/day).

Physical activity was defined as moderate or vigorous activity over the past 30 days, versus sedentary, from two questions (CDC, 2012). The first of these questions was: "Over the past 30 days did you do any vigorous activities for at least 10 min that caused heavy sweating, or large increase in breathing or heart rate? Some examples are running, lap swimming, aerobics classes or fast bicycling." The second physical activity question was: "Over the past 30 days did you do moderate activities for at least 10 min that cause only light sweating or a slight to moderate increase in breathing or heart rate? Some examples are brisk walking, bicycling for pleasure, golf, and dancing." An individual was considered to have a family history of diabetes if one of their parents, grandparents, brothers or sisters had diabetes. Body mass index and waist circumference were used as control variables because these chemicals are lipophilic.

We used SAS version 9.2 for all analyses (SAS Institute Inc., 2012). The survey logistic procedure was used for logistic regressions to allow us to make appropriate estimates from the complex sample design used in the NHANES. The "normal" group (A1c <5.7%, Table 2) was used as the reference for all logistic regressions. Our analysis incorporated both the stratification and clustering aspects of the sampling design. The proper weighting procedures include adjustments for nonresponse and post stratification. Since minorities were oversampled and a complex sampling design was employed, sampling weights provided by the NHANES for the dioxins subsample were used to compute population estimates based on weighed parameter estimates and standard errors (CDC, 2012).

Table 3
Association of 8 dioxins and dioxin-like compounds with pre-diabetes state [a].

	Pre-diabetes with A1c 5.7–5.8%		Pre-diabetes with A1c 5.9–6.4%	
	Odds ratio	95% CI	Odds ratio	95% CI
1,2,3,6,7,8-HxCDD (fg/g)				
<88.60	1.00	–	1.00	–
88.60–299.45	0.88	0.49–1.59	1.00	0.52–1.95
≥299.46	1.41	0.62–3.21	1.42	0.63–3.20
1,2,3,4,6,7,8-HpCDD (fg/g)				
<157.33	1.00	–	1.00	–
157.33–367.39	1.05	0.64–1.72	0.79	0.46–1.36
≥367.40	1.20	0.75–1.94	1.18	0.63–2.22
1,2,3,4,6,7,8,9-OCDD (fg/g)				
<1945.82	1.00	–	1.00	–
1945.82–2908.84	1.53	0.95–2.47	1.47	0.81–2.67
≥2908.85	1.05	0.69–1.61	1.46	0.87–2.45
2,3,4,7,8-PeCDF (fg/g)				
<51.12	1.00	–	1.00	–
51.12–51.75	<0.001[b]	–	1.49	0.27–8.25
≥51.76	1.05	0.73–1.53	1.56	0.96–2.55
PCB 126 (fg/g)				
<86.48	1.00	–	1.00	–
86.48–213.60	1.39	0.89–2.18	1.98	0.97–4.06
≥213.61	1.14	0.68–1.89	2.88	1.36–6.09
PCB 169 (fg/g)				
<114.98	1.00	–	1.00	–
114.98–167.16	1.25	0.68–2.30	1.48	0.76–2.86
≥167.17	0.80	0.42–1.51	1.27	0.62–2.61
PCB 118 (ng/g)				
<0.041	1.00	–	1.00	–
0.041–0.089	1.59	0.98–2.56	2.25	1.28–3.95
≥0.090	1.05	0.52–2.14	2.30	1.23–4.30
PCB 156 (ng/g)				
<0.048	1.00	–	1.00	–
0.048–0.055	1.35	0.69–2.67	1.92	0.83–4.41
≥0.056	0.76	0.36–1.60	1.31	0.70–2.44

[a] Adjusted for age, gender, race/ethnicity, education, poverty income ratio, body mass index, waist circumference, energy adjusted fruit and vegetable consumption, physical activity and family history of diabetes.
[b] No cases in this concentration range.

3. Results and discussion

A total of 11.2% of our sample had pre-diabetes, and 7.5% had total diabetes (Table 2). Pre-diabetes with A1c 5.7–5.8% was not associated with any of the 8 individual chemicals investigated (Table 3), whereas pre-diabetes with A1c 5.9–6.4% was associated with 3,3′,4,4′,5-pentachlorobiphenyl (PCB 126) and 2,3′,4,4′,5-pentachlorobiphenyl (PCB 118). Total diabetes was associated with 6 of the 8 compounds tested (Table 4). The chemicals associated with total diabetes included 1,2,3,6,7,8-hexachlorodibenzo-*p*-dioxin (HxCDD), 1,2,3,4,6,7,8,9-octachlorodibenzo-*p*-dioxin (OCDD), 2,3,4,7,8-pentachlorodibenzofuran (PeCDF), 3,3′,4,4′,5,5′-hexachlorobiphenyl (PCB 169), PCB 118, and 2,3,3′,4,4′,5-hexachlorobiphenyl (PCB 156).

As with the 8 individual chemicals tested, there was no relationship of the number of compounds elevated with A1c 5.7–5.8% (Table 5). However, pre-diabetes with A1c 5.9–6.4% was associated with 1–13 compounds elevated. Overall, the odds

Table 4
Association of 8 dioxins and dioxin-like compounds with diabetes, and diabetes plus A1c ≥5.9%[a].

	Total diabetes		Diabetes plus A1c ≥5.9%	
	Odds ratio	95% CI	Odds ratio	95% CI
1,2,3,6,7,8-HxCDD (fg/g)				
<88.60	1.00	–	1.00	–
88.60–299.45	1.21	0.67–2.18	1.12	0.73–1.71
≥299.46	2.25	1.16–4.37	1.89	1.22–2.91
1,2,3,4,6,7,8-HpCDD (fg/g)				
<157.33	1.00	–	1.00	–
157.33–367.39	1.34	0.65–2.74	1.06	0.64–1.77
≥367.40	1.73	0.88–3.40	1.52	0.94–2.44
1,2,3,4,6,7,8,9-OCDD (fg/g)				
<1945.82	1.00	–	1.00	–
1945.82–2908.84	1.43	0.86–2.41	1.57	1.04–2.40
≥2908.85	1.82	1.32–2.51	1.70	1.29–2.24
2,3,4,7,8-PeCDF (fg/g)				
<51.12	1.00	–	1.00	–
51.12–51.75	<0.001[b]	–	0.72	0.12–4.28
≥51.76	2.39	1.41–4.05	2.01	1.35–2.98
PCB 126 (fg/g)				
<86.48	1.00	–	1.00	–
86.48–213.60	1.00	0.54–1.83	1.27	0.75–2.15
≥213.61	1.81	0.94–3.48	2.17	1.38–3.41
PCB 169 (fg/g)				
<114.98	1.00	–	1.00	–
114.98–167.16	2.11	0.94–4.72	1.85	0.96–3.55
≥167.17	2.56	1.20–5.48	1.91	1.06–3.44
PCB 118 (ng/g)				
<0.041	1.00	–	1.00	–
0.041–0.089	2.47	1.36–4.49	2.44	1.59–3.74
≥0.090	3.53	1.64–7.58	2.97	1.72–5.11
PCB 156 (ng/g)				
<0.048	1.00	–	1.00	–
0.048–0.055	2.58	1.28–5.23	2.17	1.21–3.89
≥0.056	2.47	1.30–4.68	1.91	1.20–3.05

[a] Adjusted for age, gender, race/ethnicity, education, poverty income ratio, body mass index, waist circumference, energy adjusted fruit and vegetable consumption, physical activity and family history of diabetes.
[b] No cases in this concentration range.

ratio, for one or more compounds elevated and A1c 5.9–6.4%, was 2.47 (95% CI 1.51–4.06). In contrast, total diabetes was not associated with 1, 2 or 3 compounds elevated, but was associated with 4–8 chemicals elevated. The odds ratio, for 4 or more compounds elevated and total diabetes, was 2.62 (95% CI 1.29–5.31). Of particular note, ≥14 compounds elevated and total diabetes had an odds ratio of 5.56 (95% CI 1.94–15.92). Having multiple compounds elevated appeared to be important for total diabetes, whereas for pre-diabetes with A1c 5.9–6.4%, having a single compound elevated appeared most important.

Toxic equivalency ≥81.58 TEQ fg/g was associated with total diabetes (odds ratio 3.08, 95% CI 1.20–7.90), but was not associated with A1c 5.7–5.8%, or with A1c 5.9–6.4% (Table 6). Pre-diabetes with A1c 5.9–6.4% was associated with the third quartile of toxic equivalency (34.16–81.57 TEQ fg/g). The fact that the third quartile was significant and the fourth quartile was not, suggests that TEQ methodology is not well suited to evaluation of A1c 5.9–6.4%. In contrast, it does seem useful with total diabetes as the outcome.

We also tested diabetes plus A1c ≥5.9% as a possible outcome to take into consideration a larger group of people at risk for diabetes. Seven of the 8 chemicals tested individually were associated with diabetes plus A1c ≥5.9% (Table 4). The only compound among the 8 that was not associated was 1,2,3,4,6,7,8-heptachlorodibenzo-*p*-dioxin (HpCDD). For diabetes plus A1c ≥5.9%, one or more compounds elevated had an odds ratio of 1.87 (95% CI 1.25–2.81), and ≥14 chemicals elevated had an odds ratio of 3.94 (95% CI 1.60–9.67) (Table 5). Using the toxic equivalency approach, diabetes plus A1c ≥5.9% was associated with 34.16–81.57 TEQ fg/g (odds ratio 2.00, 95% CI 1.06–3.77) and with ≥81.58 TEQ fg/g (odds ratio 2.48, 95% CI 1.21–5.11), indicating that half the population has elevated risk for this combination of conditions (Table 6).

The dioxin-like compounds, included in the TEQ methodology, induce responses in animal models that are characterized by severe weight loss, thymic atrophy, hepatotoxicity, edema, fetotoxicity, teratogenicity, reproductive toxicity, immunotoxicity, and enzyme induction. These responses are subsequent to Aryl hydrocarbon (Ah) receptor binding (Haws et al., 2006), which has been hypothesized to antagonize the peroxisome proliferator-activated

Table 5
Association of the number of compounds elevated with pre-diabetes state, diabetes, and diabetes plus A1c ≥5.9%[a].

Number of compounds elevated	Pre-diabetes with A1c 5.7–5.8%		Pre-diabetes with A1c 5.9–6.4%	
	Odds ratio	95% CI	Odds ratio	95% CI
0	1.00	–	1.00	–-
1	1.18	0.61–2.28	2.16	1.06–4.39
2	1.23	0.64–2.38	2.86	1.17–7.00
3	1.38	0.75–2.55	3.40	1.58–7.33
4–8	0.96	0.53–1.75	2.02	1.11–3.66
9–13	0.88	0.51–1.53	2.85	1.41–5.76
≥14	0.54	0.16–1.77	1.98	0.55–7.12

Number of Compounds Elevated	Total diabetes		Diabetes plus A1c ≥5.9%	
	Odds ratio	95% CI	Odds ratio	95% CI
0	1.00	–	1.00	–
1	0.63	0.28–1.44	1.16	0.63–2.13
2	0.87	0.38–2.02	1.56	0.83–2.94
3	1.71	0.74–3.99	2.39	1.26–4.53
4–8	2.33	1.10–4.92	2.18	1.31–3.63
9–13	2.89	1.46–5.72	2.92	1.81–4.71
≥14	5.56	1.94–15.92	3.94	1.60–9.67

[a] Adjusted for age, gender, race/ethnicity, education, poverty income ratio, body mass index, waist circumference, energy adjusted fruit and vegetable consumption, physical activity and family history of diabetes.

Table 6
Association of the toxic equivalency (TEQ) of dioxin and dioxin-like compounds with pre-diabetes state, diabetes, and diabetes plus A1c $\geq 5.9\%$[a].

Toxic Equivalency (TEQ fg/g)	Pre-diabetes with A1c 5.7–5.8%		Pre-diabetes with A1c 5.9–6.4%	
	Odds ratio	95% CI	Odds ratio	95% CI
<13.82	1.00	–	1.00	–
13.82–34.15	1.10	0.54–2.22	0.64	0.21–1.91
34.16–81.57	1.24	0.63–2.43	2.50	1.00–6.22
≥ 81.58	1.18	0.50–2.81	2.08	0.76–5.64
Toxic Equivalency (TEQ fg/g)	Total diabetes		Diabetes plus A1c $\geq 5.9\%$	
	Odds ratio	95% CI	Odds ratio	95% CI
<13.82	1.00	–	1.00	–
13.82–34.15	0.86	0.43–1.75	0.72	0.38–1.35
34.16–81.57	1.70	0.75–3.83	2.00	1.06–3.77
≥ 81.58	3.08	1.20–7.90	2.48	1.21–5.11

[a] Adjusted for age, gender, race/ethnicity, education, poverty income ratio, body mass index, waist circumference, energy adjusted fruit and vegetable consumption, physical activity and family history of diabetes.

receptor (PPAR) and thus contribute to the pathophysiology of diabetes (Remillard and Bunce, 2002). Our findings suggest that the TEQ methodology is well suited to examine associations between dioxins, furans, and dioxin-like PCBs and total diabetes and diabetes plus A1c $\geq 5.9\%$. These outcomes represent 7.5% and 12.8% of the US population. Moreover, using diabetes plus A1c $\geq 5.9\%$ as an outcome covers a broader range of states related to glucose homeostasis, and is associated with TEQ of dioxins and dioxin-like compounds with 50% of the US population having elevated risk. Future research is needed to further elucidate the longitudinal relationships between dioxins, furans, and dioxin-like PCBs and diabetes and pre-diabetes as assessed using the TEQ methodology.

References

American Diabetes Association (ADA), 2010. Summary of revisions for the 2010 clinical practice recommendations. Diabetes Care 33(Suppl. 1), S3.

Centers for Disease Control and Prevention (CDC), 2012. National Center for Health Statistics. National Health and Nutrition Examination Survey, NHANES 1999–2004. [cited 2012 April 2] Available from URL: 〈http://www.cdc.gov/nchs/nhanes.htm〉.

Everett, C.J., Matheson, E.M., 2010. Biomarkers of pesticide exposure and diabetes in the 1999–2004 National Health and Nutrition Examination Survey. Environ. Int 36, 398–401.

Frithsen, I.L., Everett, C.J., 2012. Roles of environmental pollution and pesticides in metabolic syndrome and diabetes: The epidemiological evidence. In: Bagchi, D., Sreejayan, N. (Eds.), Nutritional and Therapeutic Interventions for Diabetes and Metabolic Syndrome. Academic Press, London, pp. 111–124.

Haws, L.C., Su, S.H., Harris, M., DeVito, M.J., Walker, N.J., Farland, W.H., Finley, B., Birnbaum, L.S., 2006. Development of a refined database of mammalian relative potency estimates for dioxin-like compounds. Toxicol. Sci. 89, 4–30.

Lee, D.H., Lee, I.K., Steffes, M., Jacobs Jr, D.R., 2007. Extended analyses of the association between serum concentrations of persistent organic pollutants and diabetes. Diabetes Care 30, 1596–1598.

Lee, D.-H., Steffes, M.W., Sjodin, A., Jones, R.S., Needham, L.L., Jacobs Jr, D.R., 2010. Low dose of some persistent organic pollutants predicts type 2 diabetes: a nested case-control study. Environ. Health Perspect. 118, 1235–1242.

Michalek, J.E., Pavuk, M., 2008. Diabetes and cancer in veterans of Operation Ranch Hand after adjustment for calendar period, days of spraying, and time spent in Southeast Asia. J. Occup. Environ. Med 50, 330–340.

Patel, C.J., Bhattacharya, J., Butte, A., 2010. An environment-wide association study (EWAS) on type 2 diabetes mellitus. PLoS One 5, e10746.

Philibert, A., Schwartz, H., Mergler, D., 2009. An exploratory study of diabetes in a first nation community with respect to serum concentrations of *p,p'*-DDE and PCBs and fish consumption. Int. J. Environ. Res. Public Health 6, 3179–3189.

Remillard, R.B., Bunce, N.J., 2002. Linking dioxins to diabetes: epidemiology and biologic plausibility. Environ. Health Perspect. 110, 853–858.

SAS Institute Inc., 2012. SAS Version 9.2. Cary, North Carolina, USA: SAS Institute Inc.

Schisterman, E.F., Whitcomb, B.W., Louis, G.M., Louis, T.A., 2005. Lipid adjustment in the analysis of environmental contaminants and human health risks. Environ. Health Perspect. 113, 853–857.

Turyk, M., Anderson, H.A., Knobeloch, L., Imm, P., Persky, V.W., 2009a. Prevalence of diabetes and body burdens of polychlorinated biphenyls, polybrominated diphenyl ethers, and *p,p'*-diphenyldichloroethene in Great Lakes sport fish consumers. Chemosphere 75, 674–679.

Turyk, M., Anderson, H., Knobeloch, L., Imm, P., Persky, V., 2009b. Organochlorine exposure and incidence of diabetes in a cohort of Great Lakes sport fish consumers. Environ. Health Perspect. 117, 1076–1082.

Uemura, H., Arisawa, K., Hiyoshi, M., Satoh, H., Sumiyoshi, Y., Morinaga, K., Kodama, K., Suzuki, T., Nagai, M., Suzuki, T., 2008. Associations of environmental exposure to dioxins with prevalent diabetes among general inhabitants in Japan. Environ. Res. 108, 63–68.

Ukropec, J., Radikova, Z., Huckova, M., Koska, J., Kocan, A., Sebokova, E., Drobna, B., Trnovec, T., Susienkova, K., Labudova, V., Gasperikova, D., Langer, P., Klimes, I., 2010. High prevalence of prediabetes and diabetes in a population exposed to high levels of an organochlorine cocktail. Diabetologia 53, 899–906.

US Census Bureau, 2012a. Poverty Thresholds [cited 2012 April 2] Available from URL: 〈http://www.census.gov/hhes/www/poverty/data/threshld/index.html〉.

US Census Bureau, 2012b. How the Census Bureau measures poverty [cited 2012 April 2] Available from URL: 〈http://www.census.gov/hhes/www/poverty/about/overview/measure.html〉.

Van den Berg, M., Birnbaum, L.S., Denison, M., De Vito, M., Farland, W., Feeley, M., Fiedler, H., Hakansson, H., Hanberg, A., Haws, L., Rose, M., Safe, S., Schrenk, D., Tohyama, C., Tritscher, A., Tuomisto, J., Tysklind, M., Walker, N., Peterson, R.E., 2006. The 2005 World Health Organization reevaluation of human and mammalian toxic equivalency factors for dioxins and dioxin-like compounds. Toxicol. Sci. 93, 223–241.

Wang, S.L., Tsai, P.C., Yang, C.Y., Leon Guo, Y., 2008. Increased risk of diabetes and polychlorinated biphenyls and dioxins: a 24-year follow-up study of the Yucheng cohort. Diabetes Care 31, 1574–1579.