Exposure to Persistent Organic Pollutants: Relationship With Abnormal Glucose Metabolism and Visceral Adiposity

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OBJECTIVE

The contribution of persistent organic pollutants (POPs) to the pandemic of type 2 diabetes mellitus and obesity has been assumed but remains speculative. Our study aimed at investigating the relationship of POP levels with detailed markers of glucose metabolism and body composition.

RESEARCH DESIGN AND METHODS

Glucose tolerance was determined in a group of normal-weight and obese individuals. Fat distribution was assessed with abdominal computed tomography (CT) scanning, determining subcutaneous adipose tissue (SAT) and visceral adipose tissue (VAT). Selected POPs (28 polychlorinated biphenyls [PCBs] and the pesticide *p,p'*-dichlorodiphenyldichloroethylene [*p,p'*-DDE]) were measured in serum. In a subset of obese individuals undergoing bariatric surgery, POPs were also measured in adipose tissue.

RESULTS

Among obese participants, serum and adipose tissue levels of POPs were significantly correlated to glucose levels during an oral glucose tolerance test. Logistic regression using a model including age, age^2, sex, family history of diabetes, BMI, CT-VAT, smoking behavior, physical activity level score, and a POP level identified serum levels of PCB153, the sum of PCBs and *p,p'*-DDE as significant predictors of abnormal glucose tolerance (odds ratio 4.6, 4.8, and 3.4, respectively; $P < 0.05$). Adipose tissue levels of *p,p'*-DDE were also significant predictors (odds ratio 81.6; $P < 0.05$). Serum levels of PCBs were inversely related to BMI, while serum and adipose tissue levels of all POPs were positively related to the CT-VAT/SAT ratio, suggesting an important role for the visceral fat compartment in POP dynamics.

CONCLUSIONS

Our findings further sustain the theory that exposure to environmentally relevant levels of POPs may exert both a diabetogenic and obesogenic effect.

The worldwide prevalence of obesity and type 2 diabetes mellitus is increasing at an alarming rate, with over 36% of the U.S. adult population now estimated to be overweight and with diabetes affecting 7.8%. The epidemic surpasses borders, with similar estimates being reported in European and developing countries (1). Recently, it has been suggested that certain environmental factors might contribute to the development of type 2 diabetes mellitus and obesity. Among these factors are

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man-made chemicals, described as persistent organic pollutants (POPs) (2,3). POPs are a group of diverse substances, including polychlorinated biphenyls (PCBs) and organochlorine pesticides (OCPs), that are resistant to biodegradation and ubiquitously present in our environment. Humans are predominantly exposed through the consumption of contaminated food, mainly meat, fish, and dairy products (3).

In vitro and animal studies suggest at least a link between exposure to certain environmental contaminants and diabetes. In vitro, PCBs trigger insulin release, while several POPs are associated with the development of insulin resistance in an animal model (4,5). Multiple epidemiological studies associate type 2 diabetes mellitus to POP exposure. In an elderly Swedish population, POP levels were predictive for the development of type 2 diabetes mellitus (6–8). Since obesity is one of the main driving factors for the development of type 2 diabetes mellitus, the possible obesogenic capacities of POPs have also received attention. In vitro, the pesticide *p,p′*-dichlorodiphenyldichloroethylene (*p,p′*-DDE) is known to modulate the release of adipokines such as leptin, adiponectin, and resistin (9). Exposure of zebra fish to an environmentally relevant mixture of POPs induces weight gain (10). Several prospective and cross-sectional, mostly North American, studies indicate a positive relationship between serum levels of certain POPs (mainly PCBs and OCPs) and obesity estimates such as BMI and/or waist circumference (11–14).

The production of many POPs has been banned for several years, resulting in a decline of levels in humans (15). Although this seems reassuring, it is suspected that exposure of humans to even low levels of POPs might induce adverse health effects (16). Our study aimed at investigating the relationship of POP levels with estimates of both body composition and glucose metabolism.

RESEARCH DESIGN AND METHODS

Study Population

One hundred fifty-one adult obese subjects were prospectively recruited when visiting the weight management clinic of the Antwerp University Hospital between November 2009 and February 2012. Subjects were eligible candidates if their BMI exceeded 25 kg/m^2, with or without known history of type 2 diabetes mellitus. Pregnant women and patients with type 1 diabetes mellitus or an active psychiatric condition were excluded. A control group of 44 normal-weight (BMI $<$25 kg/m^2) volunteers, matched by age and sex, was recruited during the same period. This study was approved by the Ethical Committee of the Antwerp University Hospital (Belgian Registry number B30020097009) and registered at ClinicalTrials.gov (number NCT01778868). All participants provided their written informed consent.

Anthropometric Measures

Anthropometric measures were taken in the morning, with individuals in a fasting state and undressed. Height was measured to the nearest 0.5 cm. Body weight was measured with a digital scale to the nearest 0.2 kg. Waist circumference was measured at the midlevel between the lower rib margin and the iliac crest. Hip circumference was measured at the level of the trochanter major, and the waist-to-hip ratio (WHR) was calculated. Body composition was determined by bioimpedance analysis, and fat mass (FM) was calculated using the formula of Deurenberg (17). A computed tomography (CT) scan at the L4–L5 level was performed to measure the amount of visceral adipose tissue (VAT) and subcutaneous adipose tissue (SAT) as previously validated (18).

Physical Activity Level

Physical activity level was estimated using a validated questionnaire, combining estimates of energy expenditure during professional working hours and leisure time (19).

Blood Sampling

Venous blood samples were obtained in fasting state from an antecubital vein between 8:00 and 10:00 A.M. into sterile BD Vacutainer tubes. Blood samples for chemical analysis of POPs were immediately centrifuged at 2,500–3,000 rpm during 15 min. Serum was stored in glass vials at -20°C. In obese subjects without a known history of type 2 diabetes mellitus (n = 145), an oral glucose tolerance test (OGTT) with 75 g of glucose was performed with sampling at 0, 15, 30, 60, 90, 120, 150, and 180 min. HbA_{1c}, glucose, and insulin were measured at the hospital laboratory. Diabetes was classified according to the American Diabetes Association definition (20). The homeostasis model assessment was used to calculate insulin resistance (HOMA-IR) and insulin sensitivity (21). Area under the curve (AUC) for glucose and insulin was calculated using all eight sampling moments in the trapezoid method.

Fat Sampling

Among 66 individuals undergoing bariatric surgery, 53 agreed to provide adipose tissue samples; these were collected during surgery from both the visceral and subcutaneous fat compartment. Samples were stored in glass vials at -20°C until analysis.

Analyses of POPs

Analyses of POPs were performed at the Toxicology Centre, University of Antwerp. The samples were analyzed for 28 PCBs (for a full list, please see Supplementary Table 1) and *p,p′*-DDE. The analytical methods and quality assurance and quality control for the serum and fat samples have been published previously (14,22). We used both lipid weight–adjusted levels and nonlipid weight–adjusted data. To correct for lipid weight, total lipids were calculated with the formula proposed by Phillips et al. (23).

Total Body Levels of POPs

Several, but not all, studies have indicated that serum levels of POPs accurately reflect adipose tissue levels (24–26). In fact, several authors have used serum levels of POPs to estimate adipose tissue levels (24,25). We therefore estimated the total body levels of POPs from the bioimpedance-derived FM and the serum levels of POPs by multiplying the FM (expressed in grams) by the serum POP level (expressed in nanograms per gram lipid). As a control, we calculated the total body levels by using the available data of POP levels in adipose tissue in the subgroup that underwent bariatric surgery (n = 53). We detected a strong correlation ($r > 0.9$; $P < 0.0001$ for all POPs) between POP body levels derived from serum and adipose tissue POP levels.

Statistical Analysis

Statistical calculations were performed using IBM SPSS, version 20.0 (IBM SPSS, Chicago, IL). Levels below the method limit of detection were entered in the

database as 0.5*limit of detection. Because PCBs were always used as a mixture of different congeners, we summed the concentrations of all PCBs (ΣPCB). Normality of distribution was verified using the Kolmogorov–Smirnov test. All POP levels displayed a skewed distribution. After transformation (y = log [x + 1]), all POP levels were transformable to normality. To detect differences in serum and total body POP levels between obese and lean individuals, independent t tests or Mann–Whitney U tests were performed. Correlations between POP levels on one hand and markers of adiposity and glucose metabolism on the other hand were investigated. Both Pearson and Spearman rank correlation analyses were performed if appropriate. Partial correlation, correcting for BMI, was performed. In order to correct for multiple comparisons, a Bonferroni correction was applied and results were only considered significant at $P <$ 0.001. One-way ANOVA with a post hoc test was used to detect differences in serum and total body POP levels between groups with different glucose tolerance status. For the nonlipid weight–adjusted data, an independent samples Kruskal–Wallis test was performed, with between-category detection of significance using Mann–Whitney U tests. Multiple logistic regression was used to assess the impact of each individual POP on the diagnosis of abnormal glucose tolerance. Abnormal glucose tolerance was defined as the presence of diabetes or isolated impaired fasting glucose (IFG) or isolated impaired glucose tolerance (IGT) or combined IFG and glucose tolerance. The model included age, age^2, sex, family history of diabetes, BMI, CT-VAT (expressed in cm^2), smoking behavior, physical activity level score, and a serum level or total body level or adipose tissue POP level. We opted to include both age and age^2 to the model given the known exponential increase of POP levels with age. A model was created with each POP level separately. All variables were entered in the model simultaneously.

RESULTS

Study Population

One hundred ninety-five subjects were included in the study. The female/male ratio was similar in both the obese and lean subgroups, and the adipose tissue subgroup (Table 1). The mean age of the entire study population was 41 ± 13 years, with a mean BMI of 39.1 ± 5.4 kg/m^2 in the obese subgroup, versus 21.8 ± 2.1 kg/m^2 in the lean control group. As expected, the mean BMI in the obese subgroup that underwent fat sampling is slightly higher, due to the local eligibility criteria for bariatric surgery (BMI >35 kg/m^2 with comorbidities or BMI >40 kg/m^2) (see Supplementary Fig. 1). None of the individuals in the lean subgroup were diagnosed with diabetes. In the obese population, 20 individuals (13.5%) had known or newly diagnosed type 2 diabetes mellitus, 46 individuals (30.3%) were identified with isolated IGT, 3 (2%) and 7 (4.6%) participants had isolated IFG or combined IFG and IGT (IFG + IGT), respectively. The physical activity level of the lean subgroup was significantly higher (Table 1).

POP Levels

The dominant PCBs identified in both serum and adipose tissue samples in our population are PCB153, PCB138, and PCB180. Detailed information on the serum and adipose tissue levels of all measured congeners, the limit of quantification and detection frequency has been published previously (18,27). PCB153, PCB138, and PCB180 make up 60% of the total serum PCB profile and over 50% of the total adipose tissue PCB profile. Therefore, we limited further statistical analysis of individual PCBs to these three PCBs. In the analysis of *p,p'*-DDE, we detected one outlier in both serum and adipose tissue and excluded this individual from further statistical analysis.

We detected significantly ($P < 0.05$) lower serum levels in obese versus lean participants for PCB153, PCB180, and ΣPCB. No difference in serum levels of PCB138 and *p,p'*-DDE between lean and obese individuals was seen (see Table 1). As described previously (22), no significant differences were found between the absolute levels of POPs in the two sampled adipose tissue compartments. Moreover, we detected a very strong correlation between serum levels and adipose tissue levels of all POPs, with ρ ranging between 0.95 and 0.97 ($P < 0.01$) for the PCBs and ρ 0.99 ($P < 0.01$) for *p,p'*-DDE. We therefore used the serum levels of POPs, available in all individuals, to calculate the total body level of POPs, as described in RESEARCH DESIGN AND METHODS. We detected a significantly ($P < 0.01$) lower total body level in lean versus obese participants for all POPs (data not given).

Relation of POP Levels With Anthropometric Data

Most PCB serum levels, but not *p,p'*-DDE, displayed an inverse relationship with weight and BMI (Table 2). Results from bioimpedance analysis indicated this negative relationship was primarily based on an inverse relationship between FM and POP serum levels (Table 2). Correlation with waist, as a measure of abdominal adiposity, was not significant (Table 2). However, all serum POPs were positively related to the CT-VAT/SAT ratio (Table 2).

All but PCB180 total body levels of POPs displayed a positive association with weight, while only the total body level of PCB138 and *p,p'*-DDE were significantly linked to BMI (Table 2). All total body levels of POPs were significantly associated with waist and CT-VAT, while all the total body levels of PCBs were significantly associated with CT-VAT/SAT (Table 2).

The correlation analyses were repeated with nonlipid weight–adjusted data, which yielded very similar results (Supplementary Table 3). Correlation analyses were also repeated in the lean and obese subjects separately. (Supplementary Tables 4 and 5). In general, significances are lost, but this is probably due to a lower sample size, particularly in the lean subgroup.

Analysis of the POP levels in adipose tissue (n = 53) revealed a positive correlation between PCB levels in adipose tissue and WHR, while all POPs correlated positively with CT-VAT and CT-VAT/SAT (Table 2).

Relation of POP Levels With Glucose Metabolism

All patients on diabetes medication (n = 9 in the entire population, n = 7 in the adipose tissue group) were excluded from the correlation analyses.

Fasting glucose was positively related to serum levels of PCB153, PCB180, and ΣPCB (Table 2). In patients undergoing a full OGTT (n = 145), glucose level at 120 min during OGTT and AUC glucose correlated positively with serum levels of all POPs. Serum levels of PCB180 were

Table 1—Clinical characteristics of the total study population, the lean and the obese subgroup, and the obese subgroup with adipose tissue sampling

	Total study population (n = 195)	Lean subgroup (n = 44)	Obese subgroup (n = 151)	P	Obese subgroup with fat sampling (n = 53)
Male/female	58/137 (30/70)	13/31 (30/70)	45/106 (30/70)	NS[a]	18/35 (34/66)
Age, years	41 (18–84)	43 (19–59)	41 (18–84)	NS[b]	40 (18–58)
BMI, kg/m^2	35.2 (17.5–62.3)	21.8 (17.5–25.3)	39.1 (26.2–62.3)	<0.01[b]	42.2 (35.6–51.4)
Waist, cm	108 (61–150)	76 (61.5–94)	117 (80–150)	<0.01[b]	123 (98–150)
WHR	0.91 (0.65–1.32)	0.77 (0.65–0.96)	0.95 (0.67–1.32)	<0.01[b]	0.97 (0.73–1.21)
CT-VAT, cm^2	160 (15–481)	59 (15–195)	190 (49–481)	<0.01[c]	213 (51–481)
CT-SAT, cm^2	522 (53–1,055)	192 (53–369)	617 (282–1,055)	<0.01[b]	684 (329–1,055)
CT-VAT/SAT	0.33 (0.07–1.12)	0.33 (0.10–0.97)	0.33 (0.07–1.12)	NS[c]	0.33 (0.07–0.90)
Normal glucose tolerance	119 (61.0)	44 (100)	75 (50.0)		24 (45.3)
Isolated IGT[d]	46 (23.5)		46 (30.4)		17 (32.1)
Isolated IFG	3 (1.5)	0 (0)	3 (2.0)	<0.05[a]	1 (1.9)
IFG + IGT[d]	7 (4.0)		7 (4.6)		3 (5.7)
Diabetes	20 (10.0)	0 (0)	20 (13.0)	<0.01[a]	8 (15)
HbA_{1c}, %	5.6 (4.5–11.2)	5.2 (4.8–5.9)	5.7 (4.5–11.2)	<0.01[c]	5.9 (5.1–11.2)
HbA_{1c}, mmol/mol	38 (26–99)	33 (29–41)	39 (26–99)	<0.01[c]	41 (32–99)
Fasting glucose, mg/dL	89 (62–276)	84 (71–97)	91 (62 - 276)	NS[c]	98 (68–276)
Glucose level at 120 min during OGTT, mg/dL	145 (67–439)	NA	145 (67–439)	—	156 (67–439)
Fasting insulin, mU/L	13.7 (1.4–43.7)	5.9 (1.4–13.2)	16.0 (1.5–43.7)	<0.01[c]	18.0 (1.5–43.7)
Insulin level at 120 min during OGTT, mU/L	127.2 (1.3–1,000)	NA	127.0 (1.3–1,000)	—	123.0 (8.2–476)
HOMA-IR	3.10 (0.31–10.79)	1.23 (0.31–2.69)	3.63 (0.31–10.79)	<0.01[c]	4.35 (0.33–10.79)
Physical activity level	42.3 (19.0–64.9)	46.2 (27.0–64.9)	42.0 (19.0–64.4)	<0.01[c]	40.0 (28.0–58.6)
PCB153 serum level, ng/g lipid weight	44 (2.5–624)	62.3 (17.9–148.9)	41.6 (2.5–624)	<0.0[b]	
PCB138 serum level, ng/g lipid weight	23.3 (0.3–317)	24.8 (5.9–68.2)	23.3 (0.3–317)	NS	
PCB180 serum level, ng/g lipid weight	27.4 (1.6–432)	54.4 (9.9–158.5)	25.0 (1.6–432.0)	<0.05[b]	
ΣPCB serum level, ng/g lipid weight	170 (14.3–2,189)	242.7 (63.1–570.1)	150.9 (14.3–2,189)	<0.05[b]	
p,p′-DDE serum level, ng/g lipid weight	104.5 (8.6–3,373.0)	99.4 (19–908.6)	120.3 (8.6–3,373)	NS	

Data are presented as n (%) or mean (minimum–maximum). NA, not available; NS, not significant. [a]Differences between the obese and lean subgroup were assessed using the χ^2 test. [b]Differences between the obese and lean subgroup were assessed using the independent samples t test. [c]Differences between the obese and lean subgroup were assessed using the Mann–Whitney U test. [d]In 145 obese subjects without a known history of type 2 diabetes mellitus, an OGTT was performed.

negatively related to fasting insulin. All PCB serum levels were negatively related to homeostasis model assessment of β-cell function (HOMA-B). Total body levels of all PCBs displayed a significantly positive relation with HbA_{1c}, fasting glucose, glucose levels after 120 min, and AUC glucose. Total body levels of *p,p′*-DDE were positively related to HbA_{1c}, glucose levels after 120 min, and AUC glucose (Table 2). Adipose tissue levels of all POPs were positively associated with glucose level at 120 min during OGTT and AUC glucose (Table 2). A partial correlation, correcting for BMI, was performed with those glucose parameters displaying a normal distribution (Supplementary Table 4). The significant correlation with HOMA-B is lost, but we still detected a significant correlation with glucose levels after 120 min and AUC glucose. The correlation analyses were repeated with nonlipid weight–adjusted data, which yielded very similar results (Supplementary Table 3). Correlation analyses were also repeated in the lean and obese subjects separately. (Supplementary Tables 6 and 7). In general, significances are lost, but this is probably due to a lower sample size, particularly in the lean subgroup.

Serum levels and total body levels of POPs differed significantly between subjects with normal glucose tolerance or prediabetes (defined as isolated IFG, isolated IGT, or combined IFG + IGT) or diabetes (Table 3 and Supplementary Table 3).

Logistic regression was performed to assess the independent association of POP levels on glucose tolerance status, defined as normal versus abnormal (isolated IFG or isolated IGT or combined IFG + IGT or diabetes) (Table 4). Statistically significant models (χ^2 between 8.2 and 13.9; $P < 0.001$) are represented in Table 4. The models using lipid weight–adjusted data explained between 34.8 and 52.2% of the variance in glucose tolerance status (Table 4). As shown in Table 4, serum levels of PCB153, ΣPCB, and *p,p′*-DDE; total body levels of ΣPCB and *p,p′*-DDE; and adipose tissue levels of *p,p′*-DDE introduced in the model made a statistically significant contribution to the association with glucose tolerance. Nonlipid

Table 2—Correlation analyses between POP level anthropometric data and markers of glucose metabolism, insulin resistance, and insulin sensitivity

	Weight	BMI	FM	Fat-free mass	Waist‡	WHR‡	CT-VAT	CT-SAT‡	CT-VAT/SAT§	HbA_{1c}¶,#	Fasting glucose¶,#	Gluc 120†,#,**	AUC glucose†, #,**	Fasting insulin ‖,¶,#	Ins 120¶,#,**	AUC insulin†,#	HOMA-IR†,#	HOMA-B†,#
Serum (n = 195)																		
PCB153*	−0.24	−0.32	−0.36	−0.05	−0.13	0.09	0.14	−0.40	0.37	0.02	0.26	0.33	0.46	−0.24	0.10	0.04	−0.14	−0.40
	0.001	<0.001	<0.001	0.470	0.062	0.192	0.054	<0.001	<0.001	0.832	<0.001	<0.001	<0.001	0.001	0.270	0.642	0.052	<0.001
PCB138*	−0.10	−0.18	−0.22	0.00	−0.00	0.19	0.21	−0.27	0.36	0.09	0.25	0.30	0.42	−0.14	0.09	0.02	−0.07	−0.31
	0.143	0.012	0.002	0.952	0.986	0.006	0.003	<0.001	<0.001	0.246	0.001	<0.001	<0.001	0.055	0.311	0.830	0.357	<0.001
PCB180*	−0.30	−0.39	−0.43	−0.07	−0.20	0.04	0.08	−0.47	0.37	−0.01	0.26	0.32	0.46	−0.31	0.08	0.05	−0.19	−0.45
	<0.001	<0.001	<0.001	0.353	0.005	0.598	0.240	<0.001	<0.001	0.905	<0.001	<0.001	<0.001	<0.001	0.370	0.599	0.009	<0.001
ΣPCB*	−0.23	−0.31	−0.36	−0.05	−0.14	0.08	0.13	−0.40	0.36	0.03	0.27	0.33	0.42	−0.24	0.09	0.04	−0.14	−0.40
	0.001	<0.001	<0.001	0.475	0.057	0.249	0.069	<0.001	<0.001	0.718	<0.001	<0.001	<0.001	0.001	0.292	0.622	0.056	<0.001
p,p′-DDE*,††	−0.06	−0.07	−0.12	−0.05	0.07	0.22	0.26	−0.14	0.31	0.13	0.19	0.32	0.42	−0.07	0.13	0.01	−0.02	−0.21
	0.391	0.324	0.096	0.491	0.320	0.002	<0.001	0.046	<0.001	0.084	0.012	<0.001	<0.001	0.384	0.147	0.877	0.817	0.005
Total body level (n = 195)																		
PCB153*	0.25	0.20	0.17	0.21	0.36	0.439	0.48	0.10	0.37	0.29	0.31	0.35	0.44	0.12	0.13	0.07	0.14	−0.120
	<0.001	0.006	0.016	0.004	<0.001	<0.001	<0.001	0.147	<0.001	<0.001	<0.001	<0.001	<0.001	0.102	0.125	0.469	0.060	0.109
PCB138*	0.36	0.34	0.32	0.27	0.48	0.50	0.51	0.25	0.31	0.34	0.27	0.30	0.38	0.22	0.12	0.04	0.20	−0.30
	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	0.003	0.156	0.647	0.006	0.694
PCB180*	0.14	0.07	0.06	0.18	0.43	0.54	0.62	−0.20	0.61	0.26	0.32	0.34	0.44	0.03	0.12	0.07	0.07	−0.21
	0.324	0.625	0.387	0.013	0.001	<0.001	<0.001	0.145	<0.001	<0.001	<0.001	<0.001	<0.001	0.678	0.182	0.424	0.369	0.005
ΣPCB*	0.27	0.21	0.18	0.22	0.37	0.44	0.48	0.12	0.36	0.31	0.32	0.34	0.43	0.13	0.12	0.07	0.15	−0.11
	<0.001	0.003	0.010	0.002	<0.001	<0.001	<0.001	0.105	<0.001	<0.001	<0.001	<0.001	<0.001	0.086	0.162	0.459	0.049	0.129
p,p′-DDE*,††	0.35	0.35	0.32	0.17	0.45	0.46	0.52	0.27	0.25	0.31	0.21	0.32	0.39	0.23	0.16	0.03	0.22	0.047
	<0.001	<0.001	<0.001	0.019	<0.001	<0.001	<0.001	<0.001	0.001	<0.001	0.005	<0.001	<0.001	0.002	0.062	0.717	0.003	0.534
Adipose tissue level (n = 53)																		
PCB153†	−0.11	−0.08	−0.23	0.02	0.33	0.53	0.58	−0.35	0.63	0.44	0.39	0.52	0.63	−0.07	0.13	0.17	0.00	−0.33
	0.418	0.542	0.093	0.886	0.016	<0.001	<0.001	0.012	<0.001	0.002	0.007	<0.001	<0.001	0.659	0.390	0.301	0.981	0.028
PCB138†	−0.12	−0.05	−0.22	−0.02	0.32	0.54	0.58	−0.33	0.62	0.47	0.35	0.53	0.61	−0.03	0.13	0.15	0.04	−0.27
	0.375	0.724	0.119	0.894	0.018	<0.001	<0.001	0.016	<0.001	0.001	0.014	<0.001	<0.001	0.837	0.400	0.349	0.800	0.068
PCB180†	−0.10	−0.08	−0.24	0.11	0.35	0.55	0.60	−0.31	0.63	0.47	0.43	0.47	0.60	−0.05	0.12	0.16	0.02	−0.35
	0.478	0.536	0.083	0.451	0.011	<0.001	<0.001	0.024	<0.001	0.001	0.002	<0.001	<0.001	0.725	0.460	0.328	0.869	0.017
ΣPCB†	−0.08	−0.05	−0.21	0.06	0.36	0.53	0.59	−0.34	0.64	0.48	0.40	0.51	0.61	−0.05	0.10	0.14	0.02	−0.33
	0.563	0.700	0.129	0.645	0.008	<0.001	<0.001	0.014	<0.001	0.001	0.005	<0.001	<0.001	0.719	0.516	0.379	0.901	0.026
p,p′-DDE*,††	−0.23	−0.20	−0.22	−0.17	0.23	0.42	0.50	−0.25	0.52	0.36	0.20	0.54	0.56	0.01	0.26	0.24	0.07	−0.16
	0.102	0.889	0.121	0.235	0.098	0.002	<0.001	0.079	<0.001	0.013	0.187	<0.001	<0.001	0.941	0.093	0.139	0.646	0.294

The r value is represented on the first line and the P value on the second line. Gluc 120, glucose level at 120 min during OGTT; Ins 120, insulin level at 120 min during OGTT. ‡Variables were not normally distributed in the entire group and were normally distributed in the adipose tissue group. §Variables were not normally distributed in the entire group and were transformed to normality with square root transformation in the adipose tissue group. ¶HbA_{1c}, fasting glucose, fasting insulin, and insulin level at 120 min during OGTT were not transformable to normality in the serum and total body level group; therefore Spearman rank correlation was performed. #Patients on diabetes medication were excluded from these analyses. †Levels were transformed to normality with square root transformation. **OGTT-derived data (glucose level at 120 min during OGTT, AUC glucose, insulin level at 120 min during OGTT, AUC insulin) were available in 145 participants. ‖Levels were transformed to normality using logarithmic transformation (10 log) in the adipose tissue group. *Levels were transformed to normality using logarithmic transformation (10 log). ††For the analysis of *p,p′*-DDE, one participant was excluded because of outlier values.

Table 3—One-way between-group ANOVA with post hoc comparisons using the Tukey test

	NGT (n = 119)	Prediabetes (n = 56)	DM (n = 20)
Serum levels (ng/g lipid)			
PCB153*	39.4† (3.9–283.0)	48.7 (2.5–624.0)	90.0† (9.7–513.0)
PCB138*	20.2§,† (0.3–106.0)	28.0§ (0.3–317.0)	51.5† (6.9–263.0)
PCB180*	24.1‡ (1.6–251.0)	29.4‡ (2.1–404.0)	51.0 (5.2–432.0)
ΣPCB*	146.3§,‡ (18.6–1,146.0)	181.0§ (14.3–2,189.0)	320.7‡ (35.4–2,103.0)
p,p′-DDE*	74.4‡,† (8.6–908.6)	236.8‡ (9.5–3,373.0)	312.7† (22.2–2,395.0)
Total body levels (ng*10^{-3})			
PCB153*	1.12†,|| (0.18–11.32)	2.41† (0.15–28.69)	4.12|| (0.52–35.01)
PCB138*	0.61†,|| (0.02–6.33)	1.31† (0.02–22.57)	2.38|| (0.37–22.91)
PCB180*	0.80†,|| (0.08–10.04)	1.31† (0.13–15.63)	2.39|| (0.28–19.34)
ΣPCB*	4.25†,|| (0.63–45.84)	9.11† (0.86–120.33)	14.90|| (1.88–133.96)
p,p′-DDE*	2.40†,|| (0.29–35.25)	7.1† (0.6–170.1)	14.71|| (1.18–208.60)

Data are presented as median (minimum–maximum) of the nontransformed variables. NGT, normal glucose tolerance; Prediabetes, isolated IGT or isolated IFG or combined IFG and IGT; DM, diabetes mellitus, according to the criteria of the American Diabetes Association. *Levels were transformed to normality using logarithmic transformation (10 log). †$P < 0.001$. §$P < 0.05$. ‡$P < 0.01$. ||$P < 0.001$.

weight–adjusted serum levels of POPs were no significant contributors to the models, but the overall strength of the models was less compared with those models using the lipid weight–adjusted data. We have performed regression analysis, using tertiles or quintiles of serum levels (both lipid weight–adjusted and nonlipid weight–adjusted) and of body burden levels, but we were not able to detect any nonlinear dose response relationships.

CONCLUSIONS

Exposure to endocrine disrupting POPs has recently emerged as a potential contributor to the pandemic of both type 2 diabetes mellitus and obesity (3,28). Our study relates a range of POPs to detailed anthropometric measures and glucose and insulin levels (basal and post-glucose load). Serum levels of POPs were significantly higher in lean individuals. This is most likely explained by the dilution capabilities of POPs; since these lipophilic substances are preferentially stored in adipose tissue, a higher percentage of body fat will lead to efficient storage and a higher total body level. As such, the serum levels exhibited a negative relationship with weight and BMI, whereas total body POP levels displayed a positive relationship. Previous studies with PCBs and *p,p′*-DDE have focused on serum levels and yielded both positive and negative relationships with weight and BMI (24,29,30). Inverse

Table 4—Logistic regression analysis assessing the impact of POP serum levels, total body levels, and adipose tissue levels on the presence of abnormal glucose tolerance

Risk factor	Odds ratio	95% CI	P value	Cox and Snell R^2	Nagelkerke R^2
Serum levels (ng/g lipid)					
Model with PCB153				34.8	47.4
BMI	1.092	1.015–1.175	0.019		
CT-VAT	1.011	1.003–1.018	0.004		
PCB153	4.640	0.998–21.576	0.050		
Model with ΣPCB				35.1	47.7
BMI	1.095	1.016–1.179	0.017		
CT-VAT	1.011	1.003–1.011	0.004		
ΣPCB*	4.875	1.118–21.251	0.035		
Model with *p,p′*-DDE				35.0	47.6
BMI	1.076	1.000–1.156	0.049		
CT-VAT	1.011	1.003–1.018	0.006		
p,p′-DDE*	3.467	1.078–11.149	0.037		
Total body levels (ng*10^{-3})					
Model with ΣPCB				34.9	47.5
CT-VAT	1.010	1.003–1.018	0.006		
ΣPCB*	4.399	1.57–24.08	0.042		
Model with *p-p′*-DDE				34.6	47.1
CT-VAT	1.010	1.003–1.018	0.007		
p-p′-DDE*	3.181	1.033–9.797	0.044		
Adipose tissue levels (ng/g lipid)					
Model with *p,p′*-DDE				38.7	52.2
p,p′-DDE*	81.693	1.178–5,665.893	0.042		

Abnormal glucose tolerance was defined as the presence of diabetes or isolated IFG or isolated glucose tolerance or combined IFG and IGT. The model included age, age^2, sex, family history of diabetes, BMI, CT-VAT (expressed in cm^2), smoking behavior, physical activity level score, and a POP level. *Variables were introduced in the model after log transformation. Representation is restricted to significant contributors only.

associations are ascribed to recent exposure and ensuing dilution during uptake, whereas positive associations are attributed to a more historic, past exposure. In our study, the POP levels in adipose tissue exposed no relationship with either weight or BMI. Recent European studies detected a positive relationship between BMI and OCP adipose tissue levels (31,32). The populations in these studies included individuals with a BMI ranging from 22–34 and 21–48 kg/m^2, respectively (31,32). It is possible that the range of BMI in our group (33–51 kg/m^2) is too restricted to detect a difference according to BMI.

Central adiposity is strongly linked to cardiovascular disease and type 2 diabetes mellitus. We report a positive association between total body levels of POPs and waist circumference. As previously reported by our group (22), the link between waist and adipose tissue levels of POPs was stronger in VAT, compared with SAT, despite the fact that absolute POP levels did not differ statistically between VAT and SAT. Adipose tissue levels of POPs were positively related to the amount of CT-VAT but not significantly related to the amount of CT-SAT. Interestingly, serum and total body levels of POPs also show a clear positive correlation with the CT-VAT/SAT ratio. It therefore cannot be ruled out that the relative effect of POPs might be more pronounced in the metabolically more active visceral fat compartment. This finding strengthens our theory that the biological effects of POPs might differ between fat compartments. The only previous study to use CT data on VAT and SAT detected PCB congener-specific correlations that did not differ between adipose tissue compartments (32). However, they did not include normal-weight individuals, potentially creating a bias. Other studies, mainly focusing on serum levels of PCBs and OCPs, have reported an inverse relationship between serum levels of POPs and waist circumference (11,16,27,33). Recent animal data suggest that POP exposure influences body composition, rather than body weight per se (34).

There is growing evidence that exposure to POPs can be considered as an additional risk factor for the development of type 2 diabetes mellitus (8,35–38). Our analyses indicate indeed a positive correlation between fasting glucose, 2 h post load and AUC glucose levels on the one hand and serum and total body levels of all POPs on the other hand. Total body levels of all POPs were also significantly related to HbA_{1c}. We did not detect a relationship with fasting insulin, thus failing to identify a compensatory rise in insulin secretion. This finding seems to support the hypothesis of direct POP toxicity to the β-cell as a potential causative factor in the development of type 2 diabetes mellitus (28). The negative link between POP levels and HOMA-B, although a crude estimate of insulin secretion, seems to strengthen this finding. However, recent animal data contribute a role for insulin resistance and hyperinsulinemia in the possible pathways linking POP exposure to type 2 diabetes mellitus (39). To the best of our knowledge, we are the first group to assess the influence of POP levels on glucose metabolism in an obese population using the standardized and validated 75 g OGTT. The finding that 2-h post load glucose levels and AUC glucose levels are positively correlated with total body levels of all POPs further strengthens the hypothesis of the diabetogenic capacities of POPs in vivo. Our regression model indicated a statistically significant role for ΣPCB and *p,p'*-DDE in particular, along with known risk factors such as visceral adiposity and a positive familial history, on the risk of abnormal glucose tolerance. This finding also suggests that environmentally present levels of POPs might be able to increase the disease burden of diabetes at population level.

Although we report very detailed data on POPs and obesity and diabetes, we cannot rule out the possibility of reverse causality given the cross-sectional design of the study. Given the lipophilicity of POPs, the detected relationships might simply reflect the increased adipose mass in individuals with increased POP levels. Other chemicals of a less persistent nature, such as phthalates and their metabolites, have also been linked to obesity and disturbances of glucose metabolism but were not measured in our study (40). Because of legal restrictions for bariatric surgery in Belgium, adipose tissue sampling was only possible in a subgroup of participants, with a significantly higher BMI range. In the lean control group, all participants exhibited a normal fasting glucose. Although the presence of abnormal glucose tolerance cannot be fully excluded on this basis, the a priori chance of diagnosing abnormal glucose tolerance is small. In addition, other factors, such as diet, parity, and breastfeeding, may influence the serum levels of POPs as well. This information was, however, not collected in our study.

This study provides unique data on total body and adipose tissue levels of a range of ubiquitously present POPs. Despite some limitations, the link between POP levels and abnormal glucose tolerance further sustains the theory that environmentally relevant levels of POPs may exert a diabetogenic effect. Furthermore, the consistent positive association between POP levels and visceral adiposity is highly relevant, given the biologically detrimental effect of this fat compartment.

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