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ARTICLE

High prevalence of prediabetes and diabetes in a population exposed to high levels of an organochlorine cocktail

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Abstract
Aims/hypothesis A heavily polluted area of Eastern Slovakia was targeted by the PCBRISK cross-sectional survey to search for possible links between environmental pollution and both prediabetes and diabetes.
Methods Associations of serum levels of five persistent organic pollutants (POPs), namely polychlorinated biphenyls (PCBs), 2,2′-*bis*(4-chlorophenyl)-1,1-dichloroethylene (*p,p′*-DDE), 2,2′-*bis*(4-chlorophenyl)-1,1,1-trichloro-ethane (*p,p′*-DDT), hexachlorobenzene (HCB) and β-hexachlorocyclohexane (β-HCH), with prediabetes and diabetes were investigated in 2,047 adults. Diabetes and prediabetes were diagnosed by fasting plasma glucose in all participants and by OGTT in 1,220 compliant participants.
Results Our population was stratified in terms of individual POPs quintiles and associations between environmental pollution, prediabetes and diabetes were investigated. Prevalence of prediabetes and diabetes increased in a dose-dependent manner, with individuals in upper quintiles of individual POPs showing striking increases in prevalence of prediabetes as shown by OR and 95% CI for PCBs (2.74; 1.92–3.90), DDE (1.86; 1.17–2.95), DDT (2.48; 1.77–3.48), HCB (1.86; 1.7–2.95) and β-HCH (1.97; 1.28–3.04). Interestingly, unlike PCBs, DDT and DDE, increased levels of HCB and β-HCH seemed not to be associated with increased prevalence of diabetes. Nevertheless, individuals in the 5th quintile of the variable expressing the cumulative effect of all five POPs (sum of orders) had a more than tripled prevalence of prediabetes and more than six times higher prevalence of diabetes when compared with the 1st referent quintile.
Conclusions/interpretation Increasing serum concentrations of individual POPs considerably increased prevalence of prediabetes and diabetes in a dose-dependent manner. Interaction of industrial and agricultural pollutants in increasing prevalence of prediabetes or diabetes is likely.

Keywords Diabetes · Persistent organochlorine pollutants · Prediabetes

Abbreviations

AhR	Aryl hydrocarbon receptor
p,p′-DDE	2,2′-*bis*(4-chlorophenyl)-1,1-dichloroethylene
p,p′-DDT	2,2′-*bis*(4-chlorophenyl)-1,1,1-trichloro-ethane
FPG	Fasting plasma glucose
HCB	Hexachlorobenzene
β-HCH	β-Hexachlorocyclohexane

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PCBs	Polychlorinated biphenyls
POLL5	Variable expressing the cumulative effect of all five POPs (sum of orders)
POPs	Persistent organochlorine pollutants
TCDD	2,3,7,8-Tetrachlorodibenzo-*p*-dioxin

Introduction

Among the several adverse effects of persistent organochlorine pollutants (POPs) and other environmental toxicants (e.g. nitrate, arsenic) on human health, attention has focused on impaired glucose metabolism and the prevalence of diabetes mellitus [1]. The Air Force Health Study provided the seminal observation that spraying Agent Orange, contaminated with 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD), is associated with increased levels of fasting serum glucose [2]. That study also showed increased prevalence of diabetes, hyperinsulinaemia in fasted state and at 2 h after glucose load, and decreased insulin sensitivity in individuals with high serum TCDD levels [2–5].

Several reports based on mortality rates analysis [6, 7] or on self-reported diabetes also show associations between environmental pollution levels and prediabetes and/or diabetes [8–10]. However, in only few studies was diabetes diagnosed with the aid of fasting plasma glucose (FPG) (≥126 mg/dl [7.0 mmol/l]), HbA_{1c} (>6.1%) [9] or random glucose level (≥200 mg/dl=11.1 mmol/l) [11–13]. Our study was conducted within the PCBRISK project in an area of Eastern Slovakia that has been subjected to heavy industrial and agricultural pollution for nearly four decades. Our previous research clearly showed very high levels of polychlorinated biphenyls (PCBs), 2,2′-*bis*(4-chlorophenyl)-1,1-dichloroethylene (*p,p′*-DDE) and hexachlorobenzene (HCB) in environmental and human samples from this area [14, 15]. The same is true for levels of dioxins, furans and dioxin-like PCBs [16], as well as for OH- and $MeSO_2$-metabolites [17]. Moreover, in the population of this area increased thyroid volume, prevalence of thyroid antibodies, disruption of thyroid hormone level and pituitary–thyroid interrelations have been found [18–22]. More recently, an increase in adverse health signs has been reported in adults [23] as well as in newborns, infants and school children of this area, e.g. high levels of organochlorines in umbilical cord blood [24] and higher prevalence both of smaller thymic volume [25] and of increased hearing threshold [26].

The primary aim of this study was to carry out a cross-sectional analysis on the association of heavy industrial and agricultural pollution with prevalence of prediabetes and diabetes as diagnosed by FPG and, in more than half of the population, by OGTT.

Methods

Participants A total of 2,047 individuals (835 men, 1,212 women; age range 21–75 years) were recruited by 28 a priori selected primary care physicians from the heavily polluted east Slovakian district of Michalovce (433 men, 576 women) and from two adjacent districts of Svidnik and Stropkov (402 men, 636 women).

This was a representative sample of the population as virtually all residents have a primary care physician assigned to them on the basis of their place of residence, providing a direct link from a physician's practice to a geographical area of the district. Each of the 28 a priori selected local physicians was instructed to recruit about 60 to 100 individuals by systematic random sampling using the alphabetically ordered lists of their patients. The study was therefore able to achieve target recruitment of about 30% of participants between 21 and 40 years, and about 70% between 41 and 75 years, while keeping the ratio of men:women in the range of 40% to 60%. All participants underwent a thyroid ultrasound examination and fasting blood sampling. OGTTs were performed in 1,220 compliant individuals. Plasma glucose was analysed in the fasted state, as well as at 60 and 120 min after administration of 75 g glucose.

This investigation was conducted according to the declaration of Helsinki, approved by the Institutional Review Board and by anonymous reviewers of the European Commission. All participants provided written informed consent.

Blood samples were centrifuged within 2 h after collection in a refrigerated centrifuge. Serum and plasma aliquots were frozen, transported in a portable freezer to the laboratory and stored at −20°C until assayed.

Glucose Fasting as well as 1 and 2 h plasma glucose levels were determined using an analyser (Hitachi 901; Hitachi, Saitama-ken, Japan). Fasting plasma glucose and 2 h glucose were used to diagnose prediabetes (impaired fasting glucose: FPG >5.6 but <7.0 mmol/l and/or impaired glucose tolerance: 2 h glucose >7.8 but <11.1 mmol/l) and diabetes (FPG >7.0 mmol/l, 2 h glucose >11.1 mmol/l) according to criteria of the American Diabetes Association [27]. Present history of diabetes as diagnosed and already treated by a physician was found in 96 individuals.

Polychlorinated biphenyls and pesticides We determined 15 PCB congeners (IUPAC [International Union of Pure and Applied Chemistry] numbers 28, 52, 101, 105, 114, 118, 123, 138^{+163}, 153, 156^{+171}, 157, 167, 170, 180 and 189) and also

p,p′-DDE, 2,2′-*bis*(4-chlorophenyl)-1,1,1-trichloro-ethane (*p,p′*-DDT), HCB and β-hexachlorocyclohexane (β-HCH) in serum using a high-resolution gas chromatography device (HP 6890; Agilent, Santa Clara, CA, USA) equipped with a Ni-63 micro-electron capture detector and a 60-m DB-5 capillary column (J&W Scientific, Folsom, CA, USA) [14, 15].

For less abundant congeners, values corresponding to half the limit of detection were used. These varied from 3.9 ng/g lipid for PCB-157 to 7.5 ng/g lipid for PCB-52. For organochlorine pesticides the limits of detection were between 2.8 mg/g for γ-HCH and 7.4 ng/g for β-HCH. The sum of all 15 individual PCB congeners was calculated as the sum of PCBs including half the limit of detection for non-detected PCBs.

Enzymatic methods based on the determination of total cholesterol, non-esterified cholesterol, phospholipids and triacylglycerol were used to determine individual and total lipids in serum, and the level of organochlorines was then adjusted to the total lipid level.

Statistical evaluation To analyse the associations between serum levels of five individual POPs (PCBs, *p,p′*-DDE, HCB, *p,p′*-DDT and β-HCH) and prediabetes and/or diabetes, logistic regression analysis was used. Multivariate adjusted odds ratios were calculated with SAS Enterprise Miner 5.2 (SAS, Cary, NC, USA). Adjustable variables were age (continuous), sex and BMI (categorical). The outcome variable was either prediabetes or diabetes. Analysed POPs entered the models as categorical variables transformed to five categories (1st–5th quintiles) according to individual POPs levels. For each POP, the reference group consisted of individuals belonging to the first quintile (i.e. 20% of individuals with the lowest levels of POP). POPs entered the model either separately or simultaneously in order to identify their individual contribution to the outcome variable.

To evaluate the cumulative effects of all five POPs, we summed the ranks of each POP and the summary values were categorised by cut-off points 20th, 40th, 60th and 80th percentile values, resulting in a variable expressing the cumulative effect of all five POPs (sum of orders; POLL5).

Results

Prediabetes in individuals with increased circulating POPs Detailed phenotyping showed that our cohort consisted of 296 patients with diabetes, 973 individuals with prediabetes and 778 individuals with normal FPG. All study participants were stratified to quintiles according to levels of each individual POP. Logistic regression analysis using a stepwise factor selection showed independent association of individual POPs with prediabetes, which was independent of age, sex and BMI (Tables 1 and 2). Moreover, a gradual increase in the levels of each individual POP was associated with a stepwise increase in prevalence of prediabetes. Thus the prevalence was 69.8%, 59.9%, 82.3%, 80.8% and 72.5% higher in individuals in the 5th quintile of PCBs, HCB, *p,p′*-DDE, *p,p′*-DDT and β-HCH, respectively, when compared with the 1st (reference) quintile. Synergic interactions between individual POPs are likely, since partial correlation coefficients between *p,p′*-DDE and *p,p′*-DDT, as well as between HCB and β-HCH (0.505 and 0.369, respectively) indicated their significant association. Table 1 clearly shows that individuals in the upper three quintiles of PCBs, *p,p′*-DDE and *p,p′*-DDT showed a significantly elevated risk of being glucose-intolerant or having diabetes with respective ORs of 2.74, 2.49 and 2.48 in the 5th quintile. The same was true for the 4th and 5th quintile of HCB as well as for the 5th quintile of β-HCH (Table 1, Fig. 1).

To determine possible interactions between each of the individual POPs, the levels of all individual POPs categorised to quintiles were subjected to stepwise factor selection analysis. A significant effect of PCBs on the prevalence of prediabetes was shown, while the other pollutants were removed from the model due to low levels of significance (Table 2).

Use of the variable POLL5 enabled us to evaluate the collective effect of all five individual POPs on the prevalence of prediabetes. Interestingly, individuals in the 3rd, 4th and 5th quintiles of POLL5 had a higher probability of suffering from prediabetes than that shown for each individual POP, further indicating possible synergistic effects of industrial and agricultural pollutants (Table 1, Fig. 1).

Diabetes in individuals with increased circulating POPs Identical analyses as previously described for prediabetes were applied to determine associations of POPs with the prevalence of diabetes. They clearly showed significant associations of PCBs, *p,p′*-DDT and *p,p′*-DDE with prevalence of diabetes; associations were independent of age, sex and BMI (Table 3). Prevalence of diabetes among individuals in the 5th quintile of PCBs, HCB, *p,p′*-DDE, *p,p′*-DDT and β-HCH increased 3-, 4.1-, 4.4-, 5.5- and 4.4-fold respectively, when compared with the 1st (reference) quintile.

We further observed that individuals in the upper three quintiles of *p,p′*-DDT had a significantly elevated risk of developing diabetes, with ORs 1.84, 2.51 and 2.49 for the 3rd, 4th and 5th quintiles, respectively (Table 3). This was also true for the 4th and 5th quintiles of PCBs and for the 5th quintile of *p,p′*-DDE (Table 3), whereas levels of HCB and β-HCH adjusted for age, sex and BMI seemed not to be associated with increased prevalence of diabetes.

Table 1 Effect of different POPs on the prevalence of prediabetes

Analyte	Quintile				
	1st	2nd	3rd	4th	5th
PCBs					
In ng/g lipids	148–627	627–904	904–1,341	1,341–2,330	2,330–101,413
Cases (*n*/*n*)	189/409	211/409	257/410	291/409	321/410
Prevalence (%)	46.21	51.16	62.27	71.15	78.29
Adjusted OR (95% CI)[a]	Referent	0.99 (0.73–1.35)	1.52 (1.11–2.09)	2.27 (1.64–3.15)	2.74 (1.92–3.90)
p value	–	0.9403	0.01	<0.001	<0.001
HCB					
In ng/g lipids	21–214	214–499	499–838	838–1,364	1,364–17,927
Cases (*n*/*n*)	192/409	220/409	260/410	290/409	307/410
Prevalence (%)	46.94	53.79	63.41	70.90	74.88
Adjusted OR (95% CI)[a]	Referent	1.20 (0.86–1.68)	1.30 (0.89–1.90)	1.62 (1.07–2.46)	1.86 (1.17–2.95)
p value	–	0.2903	0.1818	0.0231	0.0083
p,p′-DDE					
In ng/g lipids	54–821	821–1,410	1,410–2,224	2,224–3,605	3,605–22,328
Cases (*n*/*n*)	175/409	226/409	265/410	284/409	319/410
Prevalence (%)	42.79	55.26	64.63	69.44	77.80
Adjusted OR (95% CI)[a]	Referent	1.30 (0.96–1.77)	1.66 (1.21–2.28)	1.93 (1.38–2.69)	2.49 (1.74–3.57)
p value	–	0.0943	0.0019	0.0001	<0.0001
p,p′-DDT					
In ng/g lipids	4–26	26–39	39–60	60–103	103–940
Cases (*n*/*n*)	177/409	220/409	267/410	285/409	320/410
Prevalence (%)	43.28	53.79	65.12	69.68	78.05
Adjusted OR (95% CI)[a]	Referent	1.17 (0.86–1.58)	1.73 (1.27–2.36)	1.88 (1.37–2.57)	2.48 (1.77–3.48)
p value	–	0.3127	0.0005	<0.0001	<0.0001
β-HCH					
In ng/g lipids	3–23	23–37	37–56	56–83	83–781
Cases (*n*/*n*)	189/409	216/409	254/408	284/411	326/410
Prevalence (%)	46.21	52.81	62.25	69.10	79.51
Adjusted OR (95% CI)[a]	Referent	1.08 (0.79–1.50)	1.25 (0.87–1.80)	1.40 (0.95–2.06)	1.97 (1.28–3.04)
p value	–	0.6299	0.2298	0.0914	0.0021
Category of POLL5					
Values (sum of orders)	5–9	6–13	14–17	18–20	21–23
Cases (*n*/*n*)	170/416	212/406	307/483	256/347	324/395
Prevalence (%)	40.87	52.2	63.56	73.78	82.03
Adjusted OR (95% CI)[a]	Referent	1.23 (0.99–1.53)	1.75 (1.41–2.16)	2.58 (2.08–3.19)	3.57 (2.88–4.42)
p value	–	0.1933	0.0007	<0.0001	<0.0001

Unless otherwise specified, values are OR and 95% CI

[a] Adjusted for age, sex and BMI

Stepwise factor selection analysis indicated that the effect of *p,p′*-DDT on the prevalence of diabetes surpasses that of other POPs, which had to be removed from the model due to low levels of significance associated with their mutual interaction (Table 2). Analysis of the cumulative effect of all five POPs revealed that individuals within the 5th quintile of the POLL5 variable had a more than 2.3-fold higher OR of having diabetes (Table 3).

Discussion

This cross-sectional study conducted in 2,047 adults, among them a considerable number exposed to high occupational and environmental pollution by PCBs and pesticides, clearly showed that highly increased serum level of five POPs was significantly associated with risk of prediabetes and diabetes in a strongly dose-dependent

Table 2 Significance of age, sex, BMI and POPs for prediabetes and diabetes

Explanatory variable	*p* value	
	Prediabetes	Diabetes
Age (years)	<0.001	<0.001
Sex (male/female)	<0.001	<0.001
BMI (kg/m^2)	<0.001	<0.001
PCBs (categorical)[a]	<0.001	–
p,p′-DDT, *p,p′*-DDE, β-HCH, HCB (categorical)[a]	>0.05	–
p,p′-DDT (categorical)[a]	–	<0.0049
PCBs, *p,p′*-DDE, β-HCH, HCB (categorical)[a]	–	>0.05
Age (years)[c]	<0.001	<0.001
Sex (male/female)[c]	<0.001	<0.001
BMI (categorical)[b,c]	<0.001	<0.001
POLL5 (categorical)[a,c]	<0.001	<0.0030

A stepwise factor selection method was used to analyse age, sex and BMI independent significance of each of the five POPs for prediabetes or diabetes. Variables were removed from the model if $p>0.05$

[a] Categories: quintile groups for each POP and for POLL5

[b] BMI categories (kg/m^2): non-obese BMI <30, obese BMI >30

[c] Analysis of the cumulative effect of all five POPs using variable POLL5 (sum of orders)

manner. This is in agreement with previous reports on abnormalities in serum insulin [2] or prevalence of self-reported diabetes [6–8, 28–30], but also with few recent large cross-sectional studies examining associations of diabetes with chronic exposure to low concentrations of POPs in the general population [11, 31, 32]. Our study strongly differs from the previous investigations in that it examined a large cohort, which had been exposed to high industrial and agricultural pollution, and showed a wide range of 5–95% POPs' levels, such as 436–5,042 ng/g lipid for a sum of 15 PCB congeners, 438–6,376 ng/g for *p,p′*-DDE and 99–2,436 ng/g for HCB (Table 1). The strength of our study also derives from the diagnosis of impaired fasting glucose/impaired glucose tolerance and diabetes, based in the majority of the population on an OGTT. While FPG was determined in all participants, an OGTT was performed in 1,220 compliant individuals (59.6% of the cohort), enabling us to study the associations of five POPs with very early changes in glucose metabolism and thus to classify participants as having diabetes and prediabetes. Interestingly, only 32% of the participants with diabetes were aware of their diabetic status.

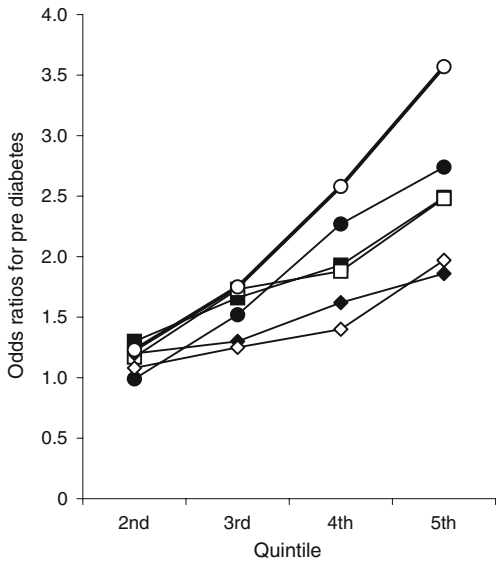

Fig. 1 The prevalence of prediabetes increases with increased circulating levels of POPs. Black circles, PCBs (15 congeners); black squares, *p,p′*-DDE; white squares, *p,p′*- DDT; black diamonds, HCB; white diamonds, β-HCH; white circles, POLL5

We and others had previously observed that presence of the major PCB congeners increases very concordantly, which jeopardised our ability to attribute any particular effect on prevalence of prediabetes or diabetes to one group of PCBs at the exclusion of the others [33]. This prompted us to use the 'whole mixture approach' in which a combination of 15 PCB congeners was investigated as if they were a single agent.

We are aware that indications of causality should be interpreted very carefully in a study with a cross-sectional design, although prospective studies have also indicated a causal relationship between environmental pollution and diabetes. Thus the Agricultural Health Study [9] examined 11,273 wives of licensed pesticide applicators, 506 (4.5%) of whom had gestational diabetes mellitus within 25 years of enrolment, while risk of diabetes was shown to be doubled in those involved in agricultural exposure during the first trimester of pregnancy. The Michigan PBB study (25 years of follow-up) showed increased incidence of diabetes in individuals with a PCB level >10.0 parts per billion (~1,300 ng/g lipid) [8]. In addition, the 24-year follow-up study of the Yucheng cohort exposed to PCBs and furans in poisoned rice oil in 1974 showed that, in women diagnosed with chloracne and thus exposed to a high level of pollutant, the OR for diabetes was 5.5 (95% CI 2.3–13.4) and that for hypertension 3.5 (1.7–7.2) as compared with those who were chloracne-free [34].

We did not find any participants with undetectable POP levels. The relatively high major POP concentrations in our referents do not therefore permit us to contribute to one of the essential questions in this field, namely whether there is a threshold level in the biological activity of individual POPs. We note that the upper limit

Table 3 Effects of different POPs on the prevalence of diabetes

Analyte	Quintile				
	1st	2nd	3rd	4th	5th
PCBs					
In ng/g lipids	148–627	627–903	904–1,341	1,341–2,330	2,330–101,413
Cases (*n*/*n*)	30/409	47/409	61/410	68/409	90/410
Prevalence (%)	7.33	11.49	14.88	16.63	21.95
Adjusted OR (95% CI)[a]	Referent	1.32 (0.77–2.26)	1.64 (0.96–2.80)	1.77 (1.05–3.02)	1.86 (1.09–3.17)
p value	–	0.3154	0.0692	0.0335	0.0223
HCB					
In ng/g lipids	21–214	214–499	499–838	838–1,364	1,364–17,927
Cases (*n*/*n*)	25/409	28/409	61/410	80/409	102/410
Prevalence (%)	6.11	6.85	14.88	19.56	24.88
Adjusted OR (95% CI)[a]	Referent	0.63 (0.33–1.17)	0.94 (0.52–1.70)	1.21 (0.66–2.23)	1.25 (0.64–2.43)
p value	–	0.1428	0.8359	0.5342	0.5154
p,p′-DDE					
In ng/g lipids	54–821	821–1,410	1,410–2,224	2,224–3,605	3,605–22,328
Cases (*n*/*n*)	23/409	43/409	67/410	61/409	102/410
Prevalence (%)	5.62	10.51	16.34	14.91	24.88
Adjusted OR (95% CI)[a]	Referent	1.43 (0.80–2.56)	1.85 (1.06–3.21)	1.34 (0.76–2.35)	1.94 (1.11–3.78)
p value	–	0.288	0.0299	0.3117	0.0198
p,p′-DDT					
In ng/g lipids	4–26	26–39	39–60	60–103	103–940
Cases (*n*/*n*)	18/409	42/409	56/410	80/409	100/410
Prevalence (%)	4.4	10.27	13.66	19.56	24.39
Adjusted OR (95% CI)[a]	Referent	1.57 (0.86–2.86)	1.84 (1.03–2.27)	2.51 (1.43–4.38)	2.49 (1.42–4.35)
p value	–	0.14254	0.0385	0.0013	0.0014
β-HCH					
In ng/g lipids	3–23	23–37	37–56	56–83	83–781
Cases (*n*/*n*)	26/409	37/409	49/408	70/411	114/410
Prevalence (%)	6.36	9.05	12.01	17.03	27.80
Adjusted OR (95% CI)[a]	Referent	0.90 (0.50–1.60)	0.77 (0.43–1.37)	0.91 (0.51–1.62)	1.08 (0.59–1.97)
p value	–	0.7135	0.3706	0.7373	0.8018
Category of POLL5					
Values (sum of orders)	5–9	10–13	14–17	18–20	21–23
Cases (*n*/*n*)	20/416	39/406	56/483	62/347	119/395
Prevalence (%)	4.81	9.61	11.59	17.87	30.13
Adjusted OR (95% CI)[a]	Referent	1.21 (0.65–2.25)	1.03 (0.59–2.02)	1.57 (0.84–2.91)	2.37 (1.27–4.39)
p value	–	0.5406	0.7836	0.1520	0.0064

Unless otherwise specified, values are OR and 95% CI

[a] Adjusted for age, sex and BMI

of PCBs of 627 ng/g lipid in our first (referent) PCBs quintile is about four times higher than that of 164 ng/g lipid reported for the 90th percentile in the NHANES Study [11]; similarly, the upper limit of *p,p′*-DDE of 821 ng/g lipid in our first quintile is still slightly higher than that of the 75th percentile (717 ng/g lipid) by the same authors. Since the selection of the reference group appears critical for the statistical evaluation, such relatively high POP levels in our referents have apparently resulted in lower ORs in the upper quintiles of individual POPs than those found in the NHANES Study [11].

Circulating lipids might significantly influence transport and tissue distribution of POPs. We previously reported significant association of PCBs, *p,p′*-DDE and HCB with circulating triacylglycerol and of *p,p′*-DDE and HCB with total cholesterol [21]. Similar an association of PCBs

(~600 ng/g) with triacylglycerol and cholesterol was recently found [35].

It has been repeatedly observed that high exposure to environmental toxicants is linked to an increased risk of diabetes. Nevertheless, molecular mechanisms explaining such associations are lacking. The most likely mechanism is one involving tissue specific up- or downregulation of gene expression, which might promote glucose intolerance and induce diabetes. It is well known that the toxic effects of several POPs are mediated via binding with aryl hydrocarbon receptor (AhR), a ligand-activated transcription factor. It has been suggested that this receptor may promote diabetogenesis by antagonising the functions of peroxisome proliferator-activated receptors, which were recently found to be linked to cellular proliferation, differentiation and apoptosis, as well as to obesity, diabetes, atherosclerosis, inflammation, cancer and ageing [1]. Vezina et al. [36] observed that transcriptional response to administration of different AhR agonists (TCDD, pentachlorodibenzofuran and PCB126) was distinctly dissimilar, indicating that different agonists have clearly specific actions. In addition, the hepatic gene expression profile for PCB153, which is not an AhR agonist, was extensive and very different from those for each of the aforementioned AhR agonists.

It has been hypothesised that the diabetogenic action of POPs could be associated with inhibition of glucose transport. In dioxin-treated mice, reduced glucose transport activity was found together with reduced copy number of the glucose transporter GLUT4 and its mRNA [37]. It was recently proposed that the ratio of *GLUT4* (also known as *SLC2A4*) mRNA to nuclear transcription factor kappa B mRNA in adipose tissue obtained from Vietnam veterans appears to be the most sensitive and reliable indicator of dioxin-induced diabetes, particularly at very low exposure levels, suggesting that the diabetogenic shift occurs in the biochemistry of adipose tissue [4]. However, a recent study of the Greenland population observed no associations between POPs and stages of glucose intolerance or markers of insulin resistance, indicating instead that POPs may affect insulin secretion [38].

Moreover, an alternative pathway has also been sought, which is predominantly focused on the association between the serum level of γ-glutamyltransferase and risk of type 2 diabetes. This was shown in a prospective cohort study in 20,158 Finnish adults followed-up for an average of 12.7 years [39] and a similar DESIR study in 5,212 French adults [40]. Results of these studies indicate that the association between serum γ-glutamyltransferase and type 2 diabetes was stronger in obesity and that obesity could not predict the risk of type 2 diabetes among participants with γ-glutamyltransferase at the low end of the normal range. This observation led to the hypothesis that the association of γ-glutamyltransferase with type 2 diabetes reflects exposure to POPs, since these reside in adipose tissue as endocrine disruptors and thus may interact with obesity to cause type 2 diabetes [41].

In conclusion, we found that increasing serum concentrations of all individual POPs were associated with progressive increase in prevalence of prediabetes and that increasing levels of PCBs, *p,p′*-DDE and *p,p′*-DDT, but not of HCB and β-HCH were associated with increased prevalence of diabetes. In addition, this study showed a synergistic effect of the five POPs in determining the prevalence of prediabetes and diabetes in the general population of Eastern Slovakia.

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Duality of interest The authors declare that there is no duality of interest associated with this manuscript.

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