



Circulating Persistent Organic Pollutants and Body Fat Distribution, Evidence from NHANES 1999-2004

Citation

Zong, Geng, Philippe Grandjean, Hongyu Wu, and Qi Sun. 2015. "Circulating Persistent Organic Pollutants and Body Fat Distribution, Evidence from NHANES 1999-2004." *Obesity* (Silver Spring, Md.) 23 (9): 1903-1910. doi:10.1002/oby.21161. <http://dx.doi.org/10.1002/oby.21161>.

Published Version

doi:10.1002/oby.21161

Permanent link

<http://nrs.harvard.edu/urn-3:HUL.InstRepos:27320473>

Terms of Use

This article was downloaded from Harvard University's DASH repository, and is made available under the terms and conditions applicable to Other Posted Material, as set forth at <http://nrs.harvard.edu/urn-3:HUL.InstRepos:dash.current.terms-of-use#LAA>

Share Your Story

The Harvard community has made this article openly available.
Please share how this access benefits you. [Submit a story](#).

[Accessibility](#)



Published in final edited form as:

Obesity (Silver Spring). 2015 September ; 23(9): 1903–1910. doi:10.1002/oby.21161.

Circulating Persistent Organic Pollutants and Body Fat Distribution, Evidence from NHANES 1999-2004

Geng Zong, Philippe Grandjean, Hongyu Wu, and Qi Sun

GZ and QS, Department of Nutrition, Harvard T.H. Chan School of Public Health, Boston, Massachusetts, USA; PG, Department of Environmental Health, Harvard T.H. Chan School of Public Health, Boston, Massachusetts, USA; PG, Institute of Public Health, University of Southern Denmark, Odense, Denmark; HW, Institute for Translational Epidemiology, Icahn School of Medicine at Mount Sinai, New York, New York, USA; QS, Channing Division of Network Medicine, Department of Medicine, Brigham and Women's Hospital and Harvard Medical School, Boston, Massachusetts, USA

Abstract

Objective—To evaluate and compare the correlations of various circulating persistent organic pollutants (POPs) with fat mass percentages (FM%) of trunk, leg, and whole body measured by Dual-energy X-ray absorptiometry.

Methods—This study included 2358 adults (≥ 20 years) in the National Health and Nutrition Examination Survey 1999-2004. Partial Pearson correlation coefficients were calculated, after adjusting for major confounders, including age, smoking status, and history of lactation and parity. Wolfe's method was used to compare correlation coefficients derived from the same participants.

Results—Twelve POPs showed significantly different correlations with fat depots in trunk and leg regions. β -hexachlorocyclohexane, heptachlorodibenzo-p-dioxin, and octachlorodibenzo-p-dioxin, and polychlorinated biphenyl (PCB)-126 showed stronger positive correlations with trunk FM% than with leg FM%, whereas PCBs with 6 chlorines were more inversely correlated with trunk FM% than leg FM%. Age-stratified analysis showed stronger inverse correlations between POPs and trunk FM% mainly in participants <40 years, whereas stronger positive correlations between POPs and trunk FM% were observed in older participants.

Conclusions—Stronger associations between POPs and trunk fat as compared to leg fat, possibly indicated a more important role of trunk fat in the pharmacokinetics of POPs, or a stronger effect of POPs, as endocrine disruptors, on trunk fat metabolism.

Keywords

Persistent organic pollutants; body fat distribution; DXA; NHANES

Users may view, print, copy, and download text and data-mine the content in such documents, for the purposes of academic research, subject always to the full Conditions of use:http://www.nature.com/authors/editorial_policies/license.html#terms

Contact Info: Q. Sun, Department of Nutrition, Harvard T.H. Chan School of Public Health, 665 Huntington Ave., Boston, MA 02115 USA. Telephone: (617) 432-7490. ; Email: qisun@hsph.harvard.edu

Disclosure: The authors declare no conflict of interest.

Author Contributions: QS designed the study. GZ analyzed data with help from QS and PG. All authors were involved in writing the paper and had final approval of the submitted and published version.

Introduction

Persistent organic pollutants (POPs) are environmentally and biologically persistent chemicals, which mainly include organochlorine pesticides, polychlorinated dibenzo-p-dioxins (PCDDs), polychlorinated dibenzofurans (PCDFs), and polychlorinated biphenyls (PCBs) (¹). Emerging evidence has suggested that POPs may contribute to the development of metabolic disorders (^{2, 3, 4}). Accordingly, several epidemiological studies have reported strong positive associations of POPs with obesity, metabolic syndrome, and diabetes (^{5, 6, 7}). By storing and releasing these chemicals to the blood stream, adipose tissue is a major source for circulating POPs. Alternatively, some POPs are known as endocrine disruptors that may mimic estrogens or inhibit androgen and thyroid functions (^{8, 9}). Such effects may affect fat accumulation and distribution (^{8, 9}). These lines of evidence indicate a plausible, but largely unexplored pathway that links adipose tissue with pathogenesis of metabolic diseases through POP exposures.

Current knowledge agrees that fat distribution is closely related to the pathology of chronic metabolic diseases (¹⁰). The accumulation of upper-body fat and visceral fat (VAT) is associated with an increased risk of these diseases, whereas lower body fat and subcutaneous fat (SAT) are associated with a preferable cardiometabolic biomarker profile (¹⁰). Some studies have examined the distribution of POPs among different fat depots. For example, Kim *et al* recently found that VAT contains higher levels of over 20 PCBs and p,p'-dichlorodiphenyl dichloroethane (p,p'-DDD), but lower p,p'-dichlorodiphenyl trichloroethane (p,p'-DDT) in a diabetes case-control study of 50 hospitalized Korean patients (¹¹). Pestana *et al* also reported higher p,p'-DDD and p,p'-dichlorodiphenyl dichloroethylene (p,p'-DDE), but lower hexachlorocyclohexane, aldrin, and dieldrin in VAT compared to those of SAT among 12 to 23 Portuguese with obesity (¹²). In contrast, Malarvannan *et al* observed similar levels of PCB138, PCB153, PCB180, and p,p'-DDE in VAT and SAT among 52 Belgians with morbid obesity (mean body mass index [BMI]=42kg/m²) (¹³). In addition, two previous studies reported similar POP levels in gluteal or femoral adipose tissue compared to those in the breast and abdominal adipose tissue (^{14, 15}). Potential differences in the associations of circulating POPs with various fat depots have also been reported, but the studies are limited in sample sizes and the findings were inconsistent (^{13, 16, 17}). Besides, none of these studies performed formal statistical tests on differences in these associations (^{13, 16, 17}).

The hypothesis that POPs may be differentially associated with fat depots in various body compartments is important to evaluate for two additional reasons. First, since fat biopsies are used for assessing longterm lipophilic pollutant exposure (¹⁸), estimated body burden of POPs by this approach might be influenced by the anatomical location of the biopsy. Likewise, the accuracy of existing physiologically based pharmacokinetic modelling for POPs might be of concern, as the calculation usually assumes that all fat tissue represents a single, uniform compartment (¹⁹). Therefore, we explicitly examined this hypothesis by comparing the correlations coefficients of POPs with trunk fat and leg fat in a large U.S. sample from the National Health and Nutrition Examination Survey (NHANES).

Methods

Study population

NHANES is a national survey on the health and nutritional status of U.S. residents. A complex sampling process based on census information was used to randomly select a nation-wide representative sample in each survey (20). Participants completed an in-home interview with trained health professionals, and were then invited to a mobile examination center for physical examinations (21). Venous blood samples were collected without fasting requirement (21). The study protocol was approved by the institutional review board at the Centers for Disease Control and Prevention (Atlanta, Georgia), and written informed consents were obtained from all participants.

The current study involved participants surveyed between 1999 and 2004, as both body composition data and detailed POP data were available in these survey cycles. Of the 31,126 U.S. residents surveyed, 2358 people were included in the analysis (see Supplemental Material, **Derivation of final analytic sample**; Supplemental Material, Figure S1).

DXA measurements

Hologic QDR 4500A fan beam x-ray bone densitometer was used (Hologic, Inc., Bedford, Massachusetts) for DXA scans (22). Hologic Discovery software (version 12.1, Hologic, Inc.) was used to analyze original scan results and to derive fat mass and lean mass. Body regions, including head, arm, trunk, and leg, were delineated manually using tools provided by the software (see Supplemental Material, **Definition of body regions**). Missing readings for DXA data were imputed five times using sequential regression multivariate imputation in the SAS-callable software package IVEware (23). All five datasets were provided, allowing analysts to incorporate the extra variability due to imputation into analyses (23). In the current study, 263 participants had 1 or more missing DXA measurements imputed (with <5% of all data points imputed). Body fat mass percentage (FM%) for the whole body and each region (trunk and legs) was calculated as fat mass divided by total mass times 100.

Persistent organic pollutants measurements

Several categories of POPs have been measured in serum from NHANES 1999-2004. These included: 1) persistent chlorinated pesticides and their pesticide metabolites, such as p,p'-DDT, p,p'-DDE, and β -hexachlorocyclohexane (β -HCH); 2) PCDDs and PCDFs, such as 1,2,3,6,7,8-hexachlorodibenzo-p-dioxin (1,2,3,6,7,8-HxCDD), 1,2,3,4,6,7,8,9-octachlorodibenzodioxin (1,2,3,4,6,7,8,9-OCDD), and 1,2,3,4,6,7,8-heptachlorodibenzofuran (1,2,3,4,6,7,8-HpCDF); and 3) PCBs, such as dioxin-like PCB126 and PCB169, and non-dioxin like PCB138 and PCB180. Laboratory procedures for POP measurement have been described elsewhere (21). Briefly, POPs were measured in serum by high-resolution gas chromatography/isotope-dilution high-resolution mass spectrometry using isotope dilution for quantification (21). The numbers of POPs measured were 49 in NHANES 1999-2000, 64 in 2001-2002, and 68 in 2003-2004 (21). We focused on those measured in at least two survey cycles for a reasonable sample size. Both original POP concentrations and lipid-adjusted concentrations (standardized to total serum lipids based on total cholesterol and triacylglycerol) were provided. In all NHANES laboratory datasets,

values below the limit of detection (LOD) were replaced with LOD divided by the square root of 2 (²⁴). In the primary analysis, we focused on the 20 POPs with 70% participants having values above LODs, for maximizing the number of POPs involved in the main analysis while maintaining a reasonable percentage of participants with values above LODs, which leaves meaningful variations in POP values for correlation analyses. We further included 26 POPs with detection rates between 30% and 69% in a secondary analysis (²⁵).

Covariates

Information on demography, lifestyle, prevalent diseases, and history of lactation and parity for women was collected using survey questionnaires during the in-person interview (²¹). Ethnicity was categorized into non-Hispanic White, non-Hispanic Black, Mexican, other Hispanic group, and other ethnic group including multi-ethnicity (²⁶). Educational attainment was categorized as high school or below, any college, and college graduate or beyond. Smoking status was classified as never smoked, past smoker, current smoker with 10 cigarettes per day, current smoker with 11 to 20 cigarettes per day, and current smoker with 21 cigarettes per day. Alcohol consumption was divided into nondrinker, 1-3 drinks/day, or 4 drinks/day. Regular moderate-to-vigorous physical activity and self-reported chronic diseases were categorized as yes or no. History of parity was self-reported number of pregnancies that resulted in successful delivery, whereas history of lactation was defined as lactation 1 months. Trained study technicians measured body weight and standing height following a standard protocol (²⁷).

Statistical analysis

Since the amount of missing data varied among POPs, we utilized non-missing data for each POP to preserve statistical power as much as possible. Natural log-transformation was performed to improve the normality of lipid-adjusted POPs and FM%. The strength of associations between lipid-adjusted POPs and FM% were evaluated by sample-weighted partial Pearson correlation coefficients. Covariate-adjustment included age (in years), gender, ethnicity, education, regular moderate-to-vigorous physical activity, smoking status, and alcohol consumption, and, for women, history of parity and history of lactation. Correlation coefficients of lipid-adjusted POPs with trunk FM% and leg FM% were then compared using Wolfe's method for the comparison of dependent correlation coefficients estimated in the same study sample. According to this method, the hypothesis that correlation between POPs and trunk FM% equals the correlation between POPs and leg FM% is equivalent to the hypothesis that correlation between POPs and (trunk FM%–leg FM%) equals zero (²⁸). Standardized z scores of trunk fat and leg fat were used in the analysis ($Z = Z_{\text{trunk FM\%}} - Z_{\text{leg FM\%}}$), to minimize the influence of between-person variation of variables. To test the significance of the correlation coefficients between POPs and Z , Fisher's z transformation and t statistics were applied and t statistics was used to evaluate the statistical significance, with the variance for the transformed correlation coefficients being $1/(n - 3)$ (²⁸).

Because of the multiple-imputation procedure, all statistics were calculated within each imputation dataset first, and then the means of these estimates were generated to derive a composite single statistical estimate (²⁹). The variance for the composite estimates were

developed by the method of Rubin and Schenker (²⁹), in which total variance accounts for both the within-imputation variance (W) and the between-imputation variance (B) according to the formula $T = W + (6/5) \times B$. W was calculated as the average of individual variance

estimates. B was calculated as $B = \sum_{i=1}^5 (Q_i - \bar{Q})^2 / 4$, where Q_i 's are the individual estimates and \bar{Q} is the mean of the 5 individual estimates. Degree of freedom was determined using the method introduced by Barnard and Rubin (³⁰). We used 47 (the number of primary sampling units minus the number of sampling strata) as the degree of freedom for complete data (³¹). To control for age and time-trend as much as possible, we also conducted age-stratified analyses (<40 years, 40 to 59 years, 60 years). Since the patterns of correlations were similar for participants aged 40 to 59 years and 60 years (See Supplemental Material, Table S1), we combined the two subsamples.

In secondary analyses, correlation coefficients between lipid-adjusted POPs and absolute fat mass were calculated with additional adjustment of body height. We also performed factor analysis among participants with all 20 POP values above LOD (n=189), and analyzed the correlation of factors (eigenvalue>1) with region fat distribution. To take into account multiple comparisons, we applied the Holm-Bonferroni method for the correction of P values. Data were analyzed in SAS, version 9.4 (SAS Institute, Inc., Cary, North Carolina).

Results

Characteristics of the study population stratified by age are shown in Table 1. In comparison with younger adults, participants aged 40 and over were more likely to be women, non-Hispanic White, non-smokers, and non-drinkers, and with higher education. Older age was also strongly associated with more trunk fat and leg fat, as well as higher serum concentrations of all POPs.

In the total population, twelve out of the 20 POPs were significantly correlated with at least one of total FM%, trunk FM%, and leg FM% after Holm-Bonferroni corrections (Table 2). The correlations with body fat varied for different POPs: β -HCH, 1,2,3,4,6,7,8-heptachlorodibenzodioxin (1,2,3,4,6,7,8-HpCDD), 1,2,3,4,6,7,8,9-OCDD, and PCB126 were positively correlated with body fat, whereas PCBs with 6 chlorines were inversely correlated with trunk and leg FM%. When we compared the correlation coefficients between trunk FM% and leg FM% for each POP, the correlations with trunk FM% were consistently stronger than those for leg FM% for β -HCH, 1,2,3,4,6,7,8- HpCDD, 1,2,3,4,6,7,8,9-OCDD, PCB126, PCB153, PCB169, PCB170, PCB180, PCB187, PCB194, PCB196, and PCB199.

Table 3 shows age-stratified partial Pearson correlation coefficients between serum POP concentrations and body fat in participants <40 years and older participants. Overall, the inverse correlations between PCBs with 6 chlorines and body fat persisted in both age groups, but the correlations for oxychlordanes, trans-nonachlor chlordanes, p,p'-DDE, β -HCH, PCDDs, and PCBs with 5 chlorines differed between age groups. Chlordanes and p,p'-DDE were inversely associated with body fat among participants <40 years, but showed null or positive correlations with body fat among participants aged 40 and above. In contrast, β -HCH, PCDDs, and PCBs with 5 chlorines tended to be positively correlated with trunk and leg FM% among participants 40 years, but not among younger participants. When

correlations of POPs with trunk FM% and leg FM% were compared, correlations for trunk FM% remained stronger than those for leg FM%. Specifically, stronger positive correlations of β -HCH, 1,2,3,4,6,7,8-HpCDD, 1,2,3,4,6,7,8,9-OCDD, PCB118, and PCB126 with trunk FM% were observed among participants aged 40 years and above, whereas stronger inverse correlations of PCBs with 6 chlorines, including PCB138, PCB153, PCB169, PCB170, PCB180, PCB187, PCB194, PCB196, and PCB199, with trunk FM% were mainly observed in participants <40 years.

Findings were similar when absolute body fat mass was modeled (Table S2). Regarding the correlation coefficients with FM% for POPs detected in 30% to 69% participants (Table S3), the correlations for trunk FM% were generally stronger than leg FM% as well, although fewer significant correlations and significant differences in correlations were observed. Consistent with differences in correlations of POPs with trunk FM% and leg FM%, we observed significant correlations between POPs and trunk/leg fat ratio for most of these pollutants, except for β -HCH (Table S4). In the factor analysis, three factors were generated with >80% variance explained overall (Table S5). Factor 1 included PCBs with 6 chlorines, factor 2 included PCDDs and PCDFs, and factor 3 included chlorinated pesticides. In consistent with our major findings, the PCB factor showed stronger inverse correlation with trunk FM%, and the PCDD and PCDF factor showed stronger positive correlation with trunk FM%, although the pesticide factor showed no differences in correlations with regional body fat depots.

Discussion

In this large, nationwide U.S. population, we found various correlation patterns between serum lipophilic POP concentrations and regional body fat distribution. The correlations of POPs with trunk fat were generally stronger than those with leg fat, especially for β -HCH, 1,2,3,4,6,7,8- HpCDD, 1,2,3,4,6,7,8,9-OCDD, PCB126, PCB153, PCB169, PCB170, PCB180, PCB187, PCB194, PCB196, and PCB199. These findings were independent of an array of demographic and lifestyle factors, including age, sex, smoking status, history of parity, and history of lactation for women, and they were observed in both young and older participants.

To our knowledge, the current analysis is among the first to comprehensively evaluate the associations between a multitude of lipophilic POPs and body fat distribution. Previously, Malarvannan *et al* found that adipose tissue concentrations of PCB138, PCB153, PCB180, and p,p'-DDE were positively associated with VAT and VAT/SAT ratio measured by abdominal computed tomography (CT) scanning, but inversely associated with SAT among 52 patients with obesity (¹³). With a larger sample size of 192 participants, the authors confirmed similar but somewhat weaker correlations of serum POPs with VAT and VAT/SAT ratio (^{13, 16}). However, their analysis did not include important confounding factors for correlations between body fat and overall POP burden, such as age, which is closely associated with body composition and POP accumulation (³²). In another analysis that was adjusted for age and other covariates, Roos *et al* found that plasma PCB118 and PCB180 were inversely associated with both VAT and SAT measured by CT among 173 postmenopausal women who were overweight/obese (¹⁷). In addition, the absolute values of

beta coefficients from linear regression models were larger for VAT than for SAT, suggesting a closer relationship between circulating POPs and VAT as compared to SAT. In another study, Roos *et al* also showed that plasma PCB169 and PCB189 were inversely associated with VAT/SAT ratio among 287 Swedes aged 70 years, but neither associations reached statistical significance after multi-comparison correction (³³). In that study, beta coefficients between POPs and VAT in the linear regression models were stronger than those for SAT, although the authors did not explicitly compare the associations of POPs with VAT and SAT. Our analysis provided more comprehensive evidence by performing formal comparisons for correlation coefficients of circulating POPs with two different fat depots, and the results suggested potential heterogeneity of regional fat depots in relation to circulating POPs.

The interpretation of stronger correlations between POPs and trunk fat must take into consideration the biological relationship between POP metabolism and adipose tissue functionality. Because adipose tissue is the primary storage site of lipophilic POPs (¹), a stronger correlation between POPs and trunk fat may suggest that trunk fat is more important in regard to the pharmacokinetics of serum POP concentrations. Upper-body fat shows higher lipogenesis and lipolysis and more sufficient exchange with blood flow (³⁴), which may favor the storage in and the release of POPs from fat in this region (¹). Accordingly, several studies have reported significantly different concentrations of POPs in VAT and SAT (^{11, 12}), although other studies did not corroborate these findings (^{13, 14, 15}). Given the abundant evidence demonstrating a strong link between upper body fat accumulation and metabolic abnormalities, our findings are compatible with the hypothesis that POPs may partially account for this association. Clearly, more research is warranted to explore the inter-relationship among body fat distribution, POP metabolism and toxicity, and metabolic diseases.

Alternatively, the observed differences in correlations of POP with various regional fat depots may suggest stronger effect of POPs on fat metabolism in the trunk area (¹). Adipocytes are under the regulation of sex hormones by expressing corresponding receptors on cell membrane. As endocrine disruptors, some POPs may interfere with the binding process and induce excessive fat accumulation (^{8, 9}). Interestingly, one study reported higher expression of steroid receptor mRNA on the membrane of adipocytes from visceral depots than subcutaneous fat in rat model, with differences between fat depots even larger than the difference between sex for the same fat depot (³⁵). Such findings suggested POPs might lead to more adipogenesis in VAT depots (³⁶). Consistent with this, a significant positive association between prenatal cord blood DDE and waist circumference has been reported among 57 girls aged 7-9 years, showing a stronger effect of POP on trunk fat accumulation during the critical developmental stage (³⁶). Apparently, further studies are warranted to provide more mechanistic insights underlying our observations, and the implications of a closer relationship between POPs and trunk fat on their pathological roles during the development of chronic diseases need to be elucidated in future investigations.

Other interesting observations were also made in the current analysis. Depending on age, both positive and negative correlations between certain POPs, such as p,p'-DDE, and body fat were observed. According to the hypothesis of Wolff *et al* (³⁷), POP concentrations will be inversely associated with body mass during exposure stage, and after 2 to 3 half-lives in

human body, the correlations will turn into the positive direction. This model seems to be able to explain the flip in the direction of correlations between p,p'-DDE and fat accumulation by age. However, for other POPs, such as PCBs with 6 chlorines, similar patterns were not observed, which may due to other factors that are closely associated with age. Age reflects diverse lifetime POP exposures among participants in different birth cohorts, and the production and release of the POPs dramatically changed over time (37). Meanwhile, age is also a significant predictor for the accumulation of adipose tissue in largely healthy populations. Therefore, older populations may have higher fat mass and percentages of body fat than younger populations (38), leading to longer retaining of POPs (37). The persistent inverse correlations between PCBs, especially those with a higher degree of halogenation, and fat depot by different age groups may be explained by the longer half-lives of PCBs with more chlorines (37). Of note, despite these potential differences in correlation patterns between age groups, stronger correlations between POPs and trunk fat were consistently observed.

Our study was based on a nationwide sample of U.S. adults. The large sample size allows for the detection of small to moderate differences in correlations of POPs with various body fat depots. The list of POPs measured in NHANES 1999-2004 is, to our knowledge, the most comprehensive to date, which enables us to evaluate and compare different POPs in regard to their correlations with adiposity. In addition, we not only controlled for essential confounding factors, such as age, sex, smoking status, and history of parity and lactation for women, but also performed age-stratified analysis to further control the strong impact of age. Meanwhile, our study is subject to some limitations. POPs were measured in serum only, and adipose tissue levels of POPs and their correlations with POP fractions in the circulation are unclear. Second, we were unable to investigate the longitudinal relationship between POP exposure and body fat accumulation over time, due to the cross-sectional design of NHANES. Thirdly, our observations may not be generalized to all NHANES participants because only a small proportion (7.6%) of total study participants was included in this analysis. The participants included in the current analysis were younger, taller, and leaner than the rest of population examined. Future studies among populations with different characteristics are warranted.

Conclusions

In conclusion, we observed stronger correlations of circulating lipophilic POPs with trunk fat than leg fat measured by DXA. These findings may have implications both on future studies that aim at characterizing POP accumulation in the lipid compartment and on interpretation of fat tissue accumulation as an endocrine disrupting effect of these substances.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

Funding: Qi Sun was supported by a career development award, R00-HL-098459, from the National Heart, Lung, and Blood Institute.

References

1. La Merrill M, Emond C, Kim MJ, Antignac JP, Le Bizec B, Clement K, et al. Toxicological function of adipose tissue: focus on persistent organic pollutants. *Environ Health Perspect.* 2013; 121:162–169. [PubMed: 23221922]
2. Remillard RB, Bunce NJ. Linking dioxins to diabetes: epidemiology and biologic plausibility. *Environ Health Perspect.* 2002; 110:853–858. [PubMed: 12204817]
3. Biswas G, Srinivasan S, Anandatheerthavarada HK, Avadhani NG. Dioxin-mediated tumor progression through activation of mitochondria-to-nucleus stress signaling. *Proc Natl Acad Sci U S A.* 2008; 105:186–191. [PubMed: 18172213]
4. Fischer LJ, Wagner MA, Madhukar BV. Potential involvement of calcium, CaM kinase II, and MAP kinases in PCB-stimulated insulin release from RINm5F cells. *Toxicol Appl Pharmacol.* 1999; 159:194–203. [PubMed: 10486306]
5. Lee DH, Lee IK, Porta M, Steffes M, Jacobs DR Jr. Relationship between serum concentrations of persistent organic pollutants and the prevalence of metabolic syndrome among non-diabetic adults: results from the National Health and Nutrition Examination Survey 1999-2002. *Diabetologia.* 2007; 50:1841–1851. [PubMed: 17624515]
6. Lee DH, Steffes MW, Sjodin A, Jones RS, Needham LL, Jacobs DR Jr. Low dose of some persistent organic pollutants predicts type 2 diabetes: a nested case-control study. *Environ Health Perspect.* 2010; 118:1235–1242. [PubMed: 20444671]
7. Lee DH, Lind PM, Jacobs DR Jr, Salihovic S, van Bavel B, Lind L. Polychlorinated biphenyls and organochlorine pesticides in plasma predict development of type 2 diabetes in the elderly: the prospective investigation of the vasculature in Uppsala Seniors (PIVUS) study. *Diabetes Care.* 2011; 34:1778–1784. [PubMed: 21700918]
8. Hatch EE, Nelson JW, Stahlhut RW, Webster TF. Association of endocrine disruptors and obesity: perspectives from epidemiological studies. *Int J Androl.* 2010; 33:324–332. [PubMed: 20113374]
9. Newbold RR, Padilla-Banks E, Jefferson WN, Heindel JJ. Effects of endocrine disruptors on obesity. *Int J Androl.* 2008; 31:201–208. [PubMed: 18315718]
10. Tchkonja T, Thomou T, Zhu Y, Karagiannides I, Pothoulakis C, Jensen MD, et al. Mechanisms and metabolic implications of regional differences among fat depots. *Cell Metab.* 2013; 17:644–656. [PubMed: 23583168]
11. Kim KS, Lee YM, Kim SG, Lee IK, Lee HJ, Kim JH, et al. Associations of organochlorine pesticides and polychlorinated biphenyls in visceral vs. subcutaneous adipose tissue with type 2 diabetes and insulin resistance. *Chemosphere.* 2014; 94:151–157. [PubMed: 24161582]
12. Pestana D, Faria G, Sa C, Fernandes VC, Teixeira D, Norberto S, et al. Persistent organic pollutant levels in human visceral and subcutaneous adipose tissue in obese individuals--depot differences and dysmetabolism implications. *Environ Res.* 2014; 133:170–177. [PubMed: 24949816]
13. Malarvannan G, Dirinck E, Dirtu AC, Pereira-Fernandes A, Neels H, Jorens PG, et al. Distribution of persistent organic pollutants in two different fat compartments from obese individuals. *Environ Int.* 2013; 55:33–42. [PubMed: 23518385]
14. Pelletier C, Imbeault P, Tremblay A. Energy balance and pollution by organochlorines and polychlorinated biphenyls. *Obes Rev.* 2003; 4:17–24. [PubMed: 12608524]
15. Rusiecki JA, Matthews A, Sturgeon S, Sinha R, Pellizzari E, Zheng T, et al. A correlation study of organochlorine levels in serum, breast adipose tissue, and gluteal adipose tissue among breast cancer cases in India. *Cancer Epidemiol Biomarkers Prev.* 2005; 14:1113–1124. [PubMed: 15894661]
16. Dirinck EL, Dirtu AC, Govindan M, Covaci A, Van Gaal LF, Jorens PG. Exposure to persistent organic pollutants: relationship with abnormal glucose metabolism and visceral adiposity. *Diabetes Care.* 2014; 37:1951–1958. [PubMed: 24963112]

17. De Roos AJ, Ulrich CM, Sjodin A, McTiernan A. Adiposity, body composition, and weight change in relation to organochlorine pollutant plasma concentrations. *J Expo Sci Environ Epidemiol*. 2012; 22:617–624. [PubMed: 22588213]
18. Kohlmeier L, Kohlmeier M. Adipose tissue as a medium for epidemiologic exposure assessment. *Environ Health Perspect*. 1995; 103 Suppl 3:99–106. [PubMed: 7635122]
19. Lipscomb JC, Haddad S, Poet T, Krishnan K. Physiologically-based pharmacokinetic (PBPK) models in toxicity testing and risk assessment. *Adv Exp Med Biol*. 2012; 745:76–95. [PubMed: 22437814]
20. National Health and Nutrition Examination Survey. Analytic Guidelines 1999–2010. Centers for Disease Control and Prevention Web site; Available from: http://www.cdc.gov/nchs/data/series/sr_02/sr02_161.pdf [accessed 1 November 2014]
21. National Health and Nutrition Examination Survey Questionnaires, Datasets, and Related Documentation. Centers for Disease Control and Prevention Web site; Available from: http://www.cdc.gov/nchs/nhanes/nhanes_questionnaires.htm [accessed 1 November 2014]
22. National Health and Nutrition Examination Survey. Body Composition Procedures Manual. Centers for Disease Control and Prevention Web site; 2008. Available from: http://www.cdc.gov/nchs/data/nhanes/nhanes_03_04/BC.pdf [accessed 1 November 2014]
23. National Health and Nutrition Examination Survey. Technical Documentation for the 1999–2004 Dual Energy X-Ray Absorptiometry (DXA) Multiple Imputation Data Files 2008. Centers for Disease Control and Prevention Web site; Available from: http://www.cdc.gov/nchs/data/nhanes/dxa/dxa_techdoc.pdf [accessed 1 November 2014]
24. How to Determine Different Limit of Detection Fill Values in NHANES Environmental Chemical Data. Centers for Disease Control and Prevention Web site; Available from: http://www.cdc.gov/nchs/tutorials/environmental/critical_issues/limitations/Task2.htm
25. Fourth National Report on Human Exposure to Environmental Chemicals. Centers for Disease Control and Prevention Web site; Available from: <http://www.cdc.gov/exposurereport/pdf/fourthreport.pdf> [accessed 1 November 2014]
26. Sun Q, van Dam RM, Spiegelman D, Heymsfield SB, Willett WC, Hu FB. Comparison of dual-energy x-ray absorptiometric and anthropometric measures of adiposity in relation to adiposity-related biologic factors. *Am J Epidemiol*. 2010; 172:1442–1454. [PubMed: 20952596]
27. National Health and Nutrition Examination Survey (NHANES) 1999–2000 Survey Operations Manuals. Centers for Disease Control and Prevention Web site; Available from: http://www.cdc.gov/nchs/nhanes/nhanes1999-2000/manuals99_00.htm [accessed 1 November 2014]
28. Rosner, B. Fundamentals of biostatistics. 7th. Brooks/Cole, Cengage Learning; Boston: 2011.
29. Rubin DB, Schenker N. Multiple Imputation for Interval Estimation from Simple Random Samples with Ignorable Nonresponse. *Journal of the American Statistical Association*. 1986; 81:366–374.
30. Barnard J, Rubin D. Miscellanea. Small-sample degrees of freedom with multiple imputation. *Biometrika*. 1999; 86:948–955.
31. Flegal KM, Shepherd JA, Looker AC, Graubard BI, Borrud LG, Ogden CL, et al. Comparisons of percentage body fat, body mass index, waist circumference, and waist-stature ratio in adults. *Am J Clin Nutr*. 2009; 89:500–508. [PubMed: 19116329]
32. Milbrath MO, Wenger Y, Chang CW, Emond C, Garabrant D, Gillespie BW, et al. Apparent half-lives of dioxins, furans, and polychlorinated biphenyls as a function of age, body fat, smoking status, and breastfeeding. *Environ Health Perspect*. 2009; 117:417–425. [PubMed: 19337517]
33. Roos V, Ronn M, Salihovic S, Lind L, van Bavel B, Kullberg J, et al. Circulating levels of persistent organic pollutants in relation to visceral and subcutaneous adipose tissue by abdominal MRI. *Obesity (Silver Spring)*. 2013; 21:413–418. [PubMed: 23532994]
34. Wronska A, Kmiec Z. Structural and biochemical characteristics of various white adipose tissue depots. *Acta Physiol (Oxf)*. 2012; 205:194–208. [PubMed: 22226221]
35. Rodriguez-Cuenca S, Monjo M, Proenza AM, Roca P. Depot differences in steroid receptor expression in adipose tissue: possible role of the local steroid milieu. *Am J Physiol Endocrinol Metab*. 2005; 288:E200–207. [PubMed: 15367392]

36. Delvaux I, Van Cauwenberghe J, Den Hond E, Schoeters G, Govarts E, Nelen V, et al. Prenatal exposure to environmental contaminants and body composition at age 7-9 years. *Environ Res.* 2014; 132:24–32. [PubMed: 24742724]
37. Wolff MS, Anderson HA, Britton JA, Rothman N. Pharmacokinetic variability and modern epidemiology--the example of dichlorodiphenyltrichloroethane, body mass index, and birth cohort. *Cancer Epidemiol Biomarkers Prev.* 2007; 16:1925–1930. [PubMed: 17932339]
38. van der Molen GW, Kooijman SA, Slob W. A generic toxicokinetic model for persistent lipophilic compounds in humans: an application to TCDD. *Fundam Appl Toxicol.* 1996; 31:83–94. [PubMed: 8998957]

Author Manuscript

Author Manuscript

Author Manuscript

Author Manuscript

Short bullet-points**What is already known?**

1. Body fat distribution and persistent organic pollutants (POPs) are both closely related to the pathology of chronic metabolic diseases.
2. Adipose tissue is the major storage site for POPs, and a potential target of POPs' detrimental effects.
3. POP biomarkers in humans are associated with different measurements of obesity.

What does this study add?

1. In a large U.S. population, we found serum POPs consistently showed stronger correlations with trunk fat than leg fat.
2. Our finding suggested a more important role of trunk fat, either as storage site or potential target, for POPs in humans.
3. These data provided possible explanations linking body fat distribution with other chronic metabolic diseases through POPs' harmful effects.

Table 1
Characteristics of study population

Variable ^a	<40 years	40 years	P value
Number of participants ^b	1001	1357	
Age, years	29.6±0.2	54.0±0.3	
Male	533 (53.6%)	674 (48.3%)	0.04
Ethnicity			<0.001
Non-Hispanic White	425 (62.6%)	685 (74.1%)	
Non-Hispanic Black	191 (10.7%)	247 (10.4%)	
Mexican	273 (12.9%)	308 (5.3%)	
Other Hispanic	46 (5.8%)	60 (5.9%)	
Other	66 (8.0%)	57 (4.3%)	
Education			0.009
High school and below	515 (43.9%)	756 (42.2%)	
Any college	306 (32.8%)	321 (28.0%)	
College graduate and above	180 (23.2%)	280 (29.8%)	
Smoking status			<0.001
Never smoked	642 (61.6%)	712 (52.4%)	
Past smoker	123 (12.6%)	409 (28.8%)	
Current smoker with 10 cigarettes per day	120 (10.7%)	82 (4.9%)	
Current smoker with 11-20 cigarettes per day	95 (12.2%)	114 (9.4%)	
Current smoker with 21 cigarettes per day	21 (2.9%)	40 (4.4%)	
Alcohol use, drinks/day			<0.001
Nondrinkers	262 (24.2%)	503 (32.3%)	
1-3 drinks/day	477 (48.8%)	703 (56.9%)	
4 drinks/day	262 (27.0%)	151 (10.8%)	
Moderate to vigorous physical activity	674 (71.8%)	752 (63.6%)	<0.001
Body mass index, kg/m ²	26.6±0.2	27.5±0.2	<0.001
Body height, cm	169.8±0.4	168.7±0.4	0.04
Number of parity for women	1.12±0.06	2.36±0.08	<0.001
Number of lactation for women	0.62±0.05	1.19±0.08	<0.001
DXA measurement			
Whole body fat mass, kg	24.6±0.4	27.5±0.3	<0.001
Whole body fat mass percentage	31.0±0.3	34.4±0.3	<0.001
Trunk fat mass, kg	11.7±0.2	13.7±0.2	<0.001
Trunk fat mass percentage	30.1±0.3	34.2±0.3	<0.001
Leg fat mass, kg	8.95±0.14	9.38±0.12	<0.001
Leg fat mass percentage	33.3±0.4	36.0±0.3	<0.001
Trunk/leg fat ratio	1.32±0.01	1.51±0.02	<0.001
Persistent organic pollutants ^c			
Chlordane			
Oxychlordane, pg/g	8.92±0.28	22.74±0.75	<0.001

Variable ^a	<40 years	40 years	P value
Trans-nonachlor Chlordane, pg/g	14.73±0.51	36.24±1.35	<0.001
p,p'-DDE, pg/g	428.60±39.38	794.26±65.22	<0.001
β-HCH, pg/g	8.82±0.90	39.33±11.75	<0.001
PCDDs and PCDFs			
1,2,3,6,7,8-HxCDD, pg/g	17.52±0.60	39.79±1.10	<0.001
1,2,3,4,6,7,8-HpCDD, pg/g	31.68±0.93	51.03±1.44	<0.001
1,2,3,4,6,7,8,9-OCDD, pg/g	239.89±8.69	419.19±12.78	<0.001
1,2,3,4,6,7,8-HpCDF, pg/g	9.87±0.82	9.84±0.32	0.04
PCBs			
PCBs with 5 Chlorines			
PCB074, pg/g	5.13±0.17	12.19±0.36	<0.001
PCB118, pg/g	6.43±0.27	15.63±0.63	<0.001
PCB126, pg/g	17.65±0.96	33.30±1.40	<0.001
PCBs with 6 Chlorines			
PCB138, pg/g	15.67±0.56	38.03±1.08	<0.001
PCB153, pg/g	21.12±0.70	55.02±1.40	<0.001
PCB169, pg/g	10.47±0.38	24.55±0.57	<0.001
PCBs with 7 Chlorines			
PCB170, pg/g	6.42±0.31	16.35±0.41	<0.001
PCB180, pg/g	14.39±0.83	42.99±1.05	<0.001
PCB187, pg/g	4.95±0.18	12.33±0.34	<0.001
PCBs with 8 Chlorines			
PCB194, pg/g	3.91±0.22	10.98±0.35	<0.001
PCB196, pg/g	3.49±0.16	8.74±0.31	<0.001
PCB199, pg/g	2.67±0.35	10.00±0.89	<0.001

Abbreviations: p,p'-DDE, p,p'-dichlorodiphenyldichloroethylene; β-HCH, β-hexachlorocyclohexane; 1,2,3,6,7,8-HxCDD, 1,2,3,6,7,8-hexachlorodibenzo-p-dioxin; 1,2,3,4,6,7,8-HpCDD, 1,2,3,4,6,7,8-heptachlorodibenzodioxin; 1,2,3,4,6,7,8,9-OCDD, 1,2,3,4,6,7,8,9-octachlorodibenzodioxin; 1,2,3,4,6,7,8-HpCDF; PCB, polychlorinated biphenyl.

^aFor continuous variables, values are presented as mean (SE). For categorical variables, numbers and weighted percentages are presented;

^bunweighted number of participants;

^cBased on non-missing participants only: Oxychlordane, n=1317; Trans-nonachlor Chlordane, n=1477; p,p'-DDE, n=1490; β-HCH, n=1468; 1,2,3,6,7,8-HxCDD, n=1981; 1,2,3,4,6,7,8-HpCDD, n=1974; 1,2,3,4,6,7,8,9-OCDD, n=1942; 1,2,3,4,6,7,8-HpCDF, n=1891; PCB074, n=2176; PCB118, n=2177; PCB126, n=1977; PCB138, n=2176; PCB153, n=2177; PCB169, n=1972; PCB170, n=2126; PCB180, n=2177; PCB187, n=2177; PCB194, n=1500; PCB196, n=1515; and PCB199, n=1509.

Partial Pearson α correlation between lipid-adjusted persistent organic pollutants with detection rate 70% and body fat percentages

Table 2

Pollutants	N	Detected rate	Total fat %	Trunk fat %	Leg fat %
Chlordane					
Oxychlordane	1317	78%	-0.06	-0.07	-0.06
Trans-nonachlor	1477	87%	-0.05	-0.05	-0.08
p,p'-DDE	1490	100%	-0.04	-0.05	-0.05
β -HCH	1468	74%	0.06	0.08 [#]	0.01
PCDDs and PCDFs					
1,2,3,6,7,8-HxCDD	1981	75%	-0.01	-0.02	-0.02
1,2,3,4,6,7,8-HpCDD	1974	89%	0.08 [*]	0.10 ^{**}	0.004
1,2,3,4,6,7,8,9-OCDD	1942	81%	0.05	0.06 [#]	0.01
1,2,3,4,6,7,8-HpCDF	1891	77%	-0.06	-0.06	-0.05
PCBs					
PCBs with 5 Chlorines					
PCB074	2176	70%	0.01	-0.004	-0.0001
PCB118	2177	73%	0.01	0.01	-0.02
PCB126	1977	81%	0.02	0.04 [#]	-0.04
PCBs with 6 Chlorines					
PCB138	2176	81%	-0.08 [*]	-0.09 [*]	-0.08 [*]
PCB153	2177	84%	-0.17 [*]	-0.17 ^{**}	-0.15 [*]
PCB169	1972	74%	-0.27 [*]	-0.26 ^{**}	-0.25 [*]
PCBs with 7 Chlorines					
PCB170	2126	75%	-0.24 [*]	-0.24 ^{**}	-0.20 [*]
PCB180	2177	83%	-0.28 [*]	-0.28 ^{**}	-0.23 [*]
PCB187	2177	70%	-0.19 [*]	-0.20 ^{**}	-0.16 [*]
PCBs with 8 Chlorines					
PCB194	1500	72%	-0.27 [*]	-0.28 ^{**}	-0.22 [*]
PCB196	1515	70%	-0.24 [*]	-0.25 ^{**}	-0.18 [*]

Author Manuscript

Author Manuscript

Author Manuscript

Author Manuscript

Pollutants	N	Detected rate	Total fat %	Trunk fat %	Leg fat %
PCB199	1509	72%	-0.29 *	-0.29 *#	-0.24 *

Abbreviations: p,p'-DDE, p,p'-dichlorodiphenyldichloroethylene; β-HCH, β-hexachlorocyclohexane; PCDD, polychlorinated dibenzo-p-dioxins; PCDF, polychlorinated dibenzofurans; 1,2,3,6,7,8-HxCDD, 1,2,3,6,7,8-hexachlorodibenzop-dioxin; 1,2,3,4,6,7,8-HpCDD, 1,2,3,4,6,7,8-heptachlorodibenzodioxin; 1,2,3,4,6,7,8-OCDD, 1,2,3,4,6,7,8-octachlorodibenzodioxin; 1,2,3,4,6,7,8-HpCDF, PCB, polychlorinated biphenyl.

^dAll correlating variables were log-transformed. Pearson correlation coefficients were adjusted for gender, age (in years), ethnicity (non-Hispanic White, non-Hispanic Black, Hispanic, or other), education (high school or below, any college, and college graduate or above), regular moderate to-vigorous physical activity (yes, no), smoking status (never smoked, past smoker, current smoker with 10 cigarettes per day, current smoker with 11-20 cigarettes per day, or current smoker with 21 cigarettes per day), alcohol consumption (nondrinker, 1–3 drinks/day, or >4 drinks/day), history of parity and history of lactation.

^{*}P values for the correlation coefficients between persistent organic pollutants and dual-energy x-ray absorptiometry body fat percentage <0.05 after multi-test corrections using step-down Bonferroni method of Holm.

[#]P values for comparisons of correlation coefficients for persistent organic pollutants with trunk fat percentage and leg fat percentage <0.05 after multi test corrections using step-down Bonferroni method of Holm.

Table 3
Age-stratified partial Pearson α correlation between lipid-adjusted persistent organic pollutants with detection rate 70% and body fat percentages

Pollutants	N	<40 years			N	40 years		
		Total fat %	Trunk fat %	Leg fat %		Total fat %	Trunk fat %	Leg fat %
Chlordane								
Oxychlordane	571	-0.17 *	-0.19 *	-0.15 *	746	0.06	0.05	0.02
Trans-nonachlor	645	-0.14 *	-0.14 *	-0.15 *	832	0.04	0.05	-0.01
p,p'-DDE	650	-0.21 *	-0.19 *	-0.21 *	840	0.15 *	0.13 *	0.14 *
β -HCH	642	-0.04	-0.02	-0.08	826	0.19 *	0.22 **	0.07
PCDDs and PCDFs								
1,2,3,6,7,8-HxCDD	843	-0.06	-0.07	-0.05	1138	0.02	0.01	0.01
1,2,3,4,6,7,8-HpCDD	847	0.02	0.05 #	-0.04	1127	0.12 *	0.15 **	0.04
1,2,3,4,6,7,8,9-OCDD	826	-0.03	-0.02	-0.04	1116	0.14 *	0.15 **	0.07
1,2,3,4,6,7,8-HpCDF	802	-0.09	-0.10	-0.07	1089	-0.03	-0.04	-0.03
PCBs								
PCBs with 5 Chlorines								
PCB074	929	-0.07	-0.08	-0.07	1247	0.12 *	0.12 *	0.07
PCB118	931	-0.07	-0.08	-0.08	1246	0.10 *	0.13 **	0.03
PCB126	843	-0.01	-0.01	-0.04	1134	0.04	0.08 #	-0.04
PCBs with 6 Chlorines								
PCB138	931	-0.17 *	-0.19 **	-0.14 *	1245	0.01	0.01	-0.01
PCB153	930	-0.24 *	-0.26 **	-0.20 *	1247	-0.09	-0.08	-0.10
PCB169	838	-0.31 *	-0.32 **	-0.27 *	1134	-0.23 *	-0.21 *	-0.23 *
PCBs with 7 Chlorines								
PCB170	911	-0.26 *	-0.28 **	-0.22 *	1215	-0.22 *	-0.20 *	-0.19 *
PCB180	934	-0.32 *	-0.35 **	-0.26 *	1243	-0.26 *	-0.25 **	-0.22 *
PCB187	931	-0.22 *	-0.24 **	-0.18 *	1246	-0.15 *	-0.15 *	-0.15 *
PCBs with 8 Chlorines								

Pollutants	<40 years				40 years			
	N	Total fat %	Trunk fat %	Leg fat %	N	Total fat %	Trunk fat %	Leg fat %
PCB194	635	-0.26 *	-0.28 **	-0.19 *	865	-0.36 *	-0.34 **	-0.29 *
PCB196	640	-0.24 *	-0.27 **	-0.17 *	875	-0.26 *	-0.25 **	-0.22 *
PCB199	640	-0.31 *	-0.35 **	-0.22 *	869	-0.33 *	-0.31 *	-0.29 *

Abbreviations: p,p'-DDE, p,p'-dichlorodiphenyldichloroethylene; β -HCH, β -hexachlorocyclohexane; PCDD, polychlorinated dibenzo-p-dioxins; PCDF, polychlorinated dibenzofurans; 1,2,3,6,7,8-HxCDD, 1,2,3,6,7,8-hexachlorodibenzop-dioxin; 1,2,3,4,6,7,8-HpCDD, 1,2,3,4,6,7,8-heptachlorodibenzodioxin; 1,2,3,4,6,7,8-HpCDF, PCB, polychlorinated biphenyl.

^a All correlating variables were log-transformed. Pearson correlation coefficients were adjusted for gender, age (in years), ethnicity (non-Hispanic White, non-Hispanic Black, Hispanic, or other), education (high school or below, any college, and college graduate or above), regular moderate to-vigorous physical activity (yes, no), smoking status (never smoked, past smoker, current smoker with 10 cigarettes per day, current smoker with 11-20 cigarettes per day, or current smoker with 21 cigarettes per day), alcohol consumption (nondrinker, 1-3 drinks/day, or >4 drinks/day), history of parity and history of lactation.

* P values for the correlation coefficients between persistent organic pollutants and dual-energy x-ray absorptiometry body fat percentage <0.05 after multi-test corrections using step-down Bonferroni method of Holm.

P values for comparisons of correlation coefficients for persistent organic pollutants with trunk fat percentage and leg fat percentage <0.05 after multi-test corrections using step-down Bonferroni method of Holm.