Published in final edited form as:

Environ Res. 2017 April ; 154: 311–317. doi:10.1016/j.envres.2017.01.033.

Exposure to phthalates is associated with lipid profile in peripubertal Mexican youth

Wei Perng¹, Deborah J. Watkins², Alejandra Cantoral³, Adriana Mercado-García³, John D. Meeker², Martha Maria Téllez-Rojo³, and Karen E. Peterson^{1,4,5}

¹Department of Nutritional Sciences, University of Michigan School of Public Health, Ann Arbor, MI, USA

²Department of Environmental Health Sciences, School of Public Health, University of Michigan, Ann Arbor, MI, USA

³Center for Nutrition and Health Research, National Institute of Public Health, Mexico City, MX

⁴Center for Human Growth and Development, University of Michigan, Ann Arbor, MI, USA

⁵Department of Nutrition, Harvard School of Public Health, Boston, MA, USA

Abstract

Animal models indicate that endocrine disrupting chemicals (EDCs) affect circulating lipid concentrations by interfering with hepatic fatty acid oxidation. Little is known of the relationship between EDC exposure and lipid profile in humans. We measured bisphenol A (BPA) and 9 phthalate metabolites in maternal urine collected at up to three time points during pregnancy as a measure of *in utero* exposure, and in the child's urine at 8–14 years as a measure of concurrent, peripubertal exposure among 248 participants of a Mexico City pre-birth cohort. We used linear regression to examine relations of BPA and phthalate exposure with peripubertal serum lipids, while also adjusting for child age, sex, and specific gravity. While *in utero* EDC exposure was not associated with lipid profile, higher concurrent levels of mono-3-carboxypropyl phthalate (MCPP), monoethyl phthalate (MEP), and dibutyl phthalate metabolites (DBP) corresponded with lower total cholesterol and low-density lipoprotein (LDL-C) in boys; e.g., an interquartile range increment in MCPP corresponded with 7.4% (2.0%, 12.8%) lower total cholesterol and 12.7% (3.8%, 21.6%) lower LDL-C. In girls, higher urinary di-2-ethylhexyl phthalate metabolites (ΣDEHP) correlated with lower LDL-C (−7.9% [−15.4%, −0.4%]). Additional longitudinal research is needed to determine whether these associations persist beyond adolescence.

Keywords

bisphenol A (BPA); phthalates; lipids; pediatric population; peripuberty

Corresponding author: Wei Perng (perngwei@umich.edu), 1415 Washington Heights, Room 1860 SPH 1, Ann Arbor, MI 48109-2029.

STATEMENT OF INTEREST: None of the authors have any conflicts of interest.

1. INTRODUCTION

Reflecting the childhood obesity epidemic, dyslipidemia – a leading cardiovascular risk factor characterized by high circulating triglycerides, total cholesterol, and low-density lipoprotein (LDL-C), and low high-density lipoprotein cholesterol (HDL-C) – affects approximately 20% of children and adolescents in the U.S. (1, 2), and up to 50% of youth in Mexico (3, 4). These statistics are alarming as pediatric dyslipidemia is a strong independent determinant of future cardiovascular and metabolic disease risk (5).

Endocrine disrupting chemicals such as bisphenol A (BPA) and phthalates, found in food packaging materials, personal care products, pesticides, and many other consumer products, have been implicated in obesity-related metabolic disturbances like chronic inflammation and dysregulated glucose homeostasis (6). Animal models have shown that *in utero* (7, 8) and concurrent (9) exposure to BPA and phthalates alter the circulating lipid profile by interfering with hepatic fatty acid metabolism. Yet, the human literature is limited to a handful of cross-sectional studies in adults that have yielded inconsistent results (10–13). Furthermore, findings from adult populations offer little preventive insight given that many cardiovascular risk factors, including dyslipidemia, are established early in the life course and continue into adulthood (14).

Metabolic processes are most vulnerable to environmental perturbations during developmental stages characterized by hormonal fluctuations and rapid maturation of organ systems – namely, the perinatal period (15, 16) and puberty (17, 18). Therefore, understanding the potential impact of EDCs during these timeframes on lipid profile will directly inform public health prevention and intervention efforts. In this study, we sought to examine relations of *in utero* and peripubertal exposure to BPA and phthalates with serum triglycerides, total cholesterol, LDL-C, and HDL-C among 248 youth 8 to 14 years of age in Mexico City, Mexico, a transitioning setting afflicted not only by high exposure to chemical toxicants (19), but also, disproportionately high rates of obesity and obesity-related metabolic aberrances (20–22). Considering that serum lipid concentrations track from childhood into adulthood (14), elucidating the potential impact of environmental toxicants on lipid profile early in the life course is critical to developing interventions that have potential to benefit long-term cardiovascular and metabolic health.

2. MATERIALS AND METHODS

2.1 Study population

This study included participants from two of three cohorts comprising the Early Life Exposure in Mexico to Environmental Toxicants (ELEMENT) Project, a longitudinal cohort study of pregnant women and their offspring in Mexico City, Mexico. Study participants were recruited from public maternity hospitals in Mexico City between 1997 and 2004 during the first trimester of pregnancy (19, 23). Mothers provided a urine sample and completed interview-based questionnaires at up to three time points across pregnancy. In 2010, we re-contacted a subset of their offspring ($n = 250$) who were then 8–14 years of age based on availability of archived prenatal biospecimens to participate in follow-up studies.

At the in-person research visits in 2010, the children filled out an interview-based questionnaire, and provided urine and serum samples. Trained research assistants measured anthropometry, including weight and height. Our study sample included 248 mother-child pairs with adequate maternal urine collected during pregnancy or child's urine during peripuberty for quantification of the EDCs, and adequate serum volume from the child during peripuberty for the serum lipid profile assays ($n = 227$ with maternal urine and peripubertal lipid data; $n = 240$ with concurrent urine and peripubertal lipid data). The institutional review boards of the Mexico National Institute of Public Health and the University of Michigan approved research protocols. Maternal informed consent and child assent were provided for all participants.

2.2 Covariates

Upon enrollment, mothers reported on age, pre-pregnancy weight and height, smoking history, and sociodemographic characteristics. At in-person research visits that took place when the children were 8–14 years, we obtained updated sociodemographic information from the mothers. A pediatrician assessed each child to determine Tanner stage for genital (boys), breast (girls), and pubic hair (both) development. For this analysis, we dichotomized pubertal status as prepubertal vs. pubertal: boys were considered pubertal if they received an assessment of Tanner stage >1 for genital or pubic hair development, and girls were considered pubertal if they received an assessment of Tanner stage >1 for breast or pubic hair development (24).

2.3 Exposures: urinary BPA & phthalate concentrations

Our exposures of interest were BPA and 9 phthalate metabolites measured in maternal urine collected during the first, second, and third trimesters of pregnancy (“*in utero*” exposure), and in child's urine at 8–14 years of age (“concurrent” or “peripubertal” exposure). All assays were carried out at NSF International (Ann Arbor, MI, USA) using high performance liquid chromatography and tandem mass-spectrometry methods described in detail elsewhere (19, 25).

We quantified concentrations of BPA and 9 phthalate metabolites including: monoethyl phthalate (MEP), mono-*n*-butyl phthalate (MnBP), mono-isobutyl phthalate (MiBP), monobenzyl phthalate (MBzP), mono-3-carboxypropyl phthalate (MCP), mono-2-ethylhexyl phthalate (MEHP), mono-2-ethyl-5-hydroxyhexyl phthalate (MEHHP), mono-2-ethyl-5-oxohexyl phthalate (MEOHP), and mono-2-ethyl-5-carboxypentyl phthalate (MECPP). These compounds are of particular interest to the ELEMENT cohort due to their known endocrine-disrupting activities and established association with other metabolic risk factors in this population (24, 26). To assess overall *in utero* exposure, we calculated geometric mean (GM) BPA and phthalate metabolite concentrations for each individual using measurements from each trimester as we have found that this approach provides a more robust measure of exposure for short-lived toxicants, like BPA and phthalates (27, 28). However, to investigate potential windows of susceptibility during the perinatal period, we also examined associations of trimester-specific EDC concentrations with serum lipids in sensitivity analyses.

For di-2-ethylhexyl phthalate metabolites (Σ DEHP), we calculated a summary score by adding the molar sums for MEHP, MEHHP, MEOHP, and MECPP. Likewise, we created a summary score for dibutyl phthalate metabolites (Σ DBP) as the molar sum of MnBP and MiBP. As we have previously done (24), we used these scores in the analysis rather than their component metabolites to reduce the possibility of false positive associations. Values below the limit of detection (LOD) were calculated as LOD/ 2. Urinary specific gravity, an indicator of urine dilution, was measured at the time of sample analysis using a handheld digital refractometer (Atago Co., Ltd., Tokyo, Japan).

2.4 Outcomes: serum lipids during peripuberty

We measured total cholesterol, triglycerides, and high density lipoprotein cholesterol (HDL-C) in peripubertal fasting serum samples (mg/dL) using a biochemical analyzer (Cobas Mira Plus, Roche Diagnostics), and calculated low density lipoprotein cholesterol (LDL-C) as: *Total cholesterol – HDL-C – (Triglycerides/5)*. We also considered assessing the prevalence of dyslipidemia using proposed cut-offs for pediatric populations (2), but due to the small number of children with abnormal lipid levels ($n = 15$ for total cholesterol ≥ 200 mg/dL; $n = 50$ for triglycerides ≥ 100 for children <9 years or ≥ 130 mg/dL for children 10–19 years; $n = 4$ for LDL-C ≥ 130 mg/dL, and $n = 10$ for HDL-C < 40 mg/dL) (2), we examined the lipids continuously.

2.5 Statistical analysis

First, we examined the distribution of each lipid across categories of sociodemographic and perinatal characteristics to identify potential confounders. For ordinal variables, like categories of maternal age at enrollment, maternal education, and child's BMI z-score during peripuberty, we conducted a test for linear trend where the ordinal indicator for the characteristic of interest was entered into the model as a continuous variable. For binary variables, such as mother's marital status, offspring sex, and offspring pubertal status, we used the Wald test to assess for a pairwise difference. Due to non-normal distributions, we natural log-(ln)-transformed all BPA and phthalate metabolites, as well as serum HDL-C and triglycerides. Next, to evaluate for non-linear associations, we examined associations of quartiles of urinary concentrations of maternal and peripubertal urinary BPA and phthalate metabolites with each lipid using linear regression. Since associations were monotonic across quartiles, we assessed phthalates and BPA continuously to maximize power, and present results as a % difference in each lipid per interquartile range (IQR) increment in the EDC of interest as we have done in prior publications (24). In multivariable analyses, we adjusted for urinary specific gravity and child's sex and age, followed by further adjustment for pubertal status. Accounting for body mass index (BMI) z-score according to the World Health Organization (WHO) growth reference (29) did not markedly change the results, thus we did not include it in our models because it is likely an intermediate variable on the causal pathway between EDC exposure and serum lipid concentrations and adjusting for it could introduce bias into the estimate of association. We also tested for effect modification by sex and pubertal status, and conducted stratified analysis where necessary (P -interaction < 0.05).

To assess for residual confounding by age, we carried out the analysis with age- and sex-standardized lipid z-scores using data from children 5–16 years in the National Health and

Nutrition Examination Survey (NHANES) III (30) as the reference population. Using the lipid z-scores yielded similar results (data available upon request); thus, we present findings for the original lipid variables for ease of interpretability. We performed all analyses using SAS 9.4 (Cary, NC, USA).

3. RESULTS

At the peripubertal research visit, median age was 10.0 years (range: 8.1, 14.7); 47.2% were boys. Mean $\pm$ SD total cholesterol was 155.2 ± 28.4 mg/dL, HDL-C was 58.7 ± 11.9 mg/dL, LDL-C was 79.0 ± 23.2 mg/dL, and triglycerides was 87.5 ± 44.4 mg/dL. When we parameterized lipid levels according to the American Academy of Pediatrics' proposed definition of dyslipidemia for pediatric populations (2), 6.1% had abnormal total cholesterol (≥ 200 mg/dL), 16.5% had abnormal triglycerides (≥ 100 mg/dL for children <9 years; ≥ 130 mg/dL for children 10–19 years), 4.0% had abnormal HDL-C (<40 mg/dL); and 1.6% had abnormal LDL-C (≥ 130 mg/dL).

In bivariate analyses (Table 1), girls had on average 8.2 ± 3.6 mg/dL higher total cholesterol, 6.4 ± 2.9 mg/dL higher LDL-C, 19.9 ± 5.5 mg/dL higher triglycerides, and 2.2 ± 1.5 mg/dL lower HDL-C than boys. BMI z-score during peripuberty was positively associated with total cholesterol (P -trend = 0.003), LDL-C (P -trend = 0.0004), and triglycerides (P -trend <0.0001), and inversely associated with HDL-C (P -trend = 0.0001). Among girls, more advanced puberty was associated with lower total cholesterol, HDL-C, and LDL-C, and higher triglycerides (Table 1).

In multivariable analysis, we accounted for urinary specific gravity, as well as child's sex and age. We did not observe consistent or significant associations of geometric mean maternal urinary EDC concentrations across all of pregnancy with peripubertal lipid profile (Table 2), nor did we find evidence of effect modification by sex or pubertal status (all P -interaction >0.10). Results were similar when we considered trimester-specific BPA and phthalate concentrations (Table S1). However, we detected sex-specific associations with respect to concurrent exposure. Table 3 shows the % difference in lipids per interquartile range (IQR) increment in each EDC. In boys, MCP, MEP, and Σ DBP were each inversely related to total cholesterol and LDL-C, and MBzP was positively associated with HDL-C. For example, an IQR difference in MCP was associated with 7.4% (95% CI: 2.0%, 12.8%) lower total cholesterol and 12.7% (95% CI: 3.8%, 21.6%) lower LDL-C, with similar associations observed for MEP and Σ DBP (Table 3). On the other hand, an IQR difference in MBzP corresponded with 6.6% (95% CI: 0.2%, 13.5%) higher HDL-C. In girls, higher Σ DEHP corresponded with lower LDL-C (-7.9% [95% CI: -15.4% , -0.4%] per IQR Σ DEHP).

Adjusting for pubertal status did not materially change the direction, magnitude, or precision of results. As compared to estimates in Table 3, accounting for pubertal status in boys slightly attenuated the relations of MCP with total cholesterol (-6.2% [95% CI: -11.7% , -0.7%]) and LDL-C (-10.9% [-20.0% , -8%] per IQR MCP). Estimates for MEP remained relatively unchanged: -5.7% (-10.5% , -0.8%) for total cholesterol and -10.2% (-18.2% , -2.3%) for LDL-C. The magnitude of associations for Σ DBP with total cholesterol

(−5.4% [−10.7%, −0.1%]) and LDL-C (−7.7% [−16.6%, −1.2%]) were somewhat diminished, while the positive relationship between MBzP and HDL-C was slightly strengthened (8.0% [1.4%, 15.1%]). In girls, the estimate for ΣDEHP and LDL-C remained the same after adjustment for pubertal status, although the upper confidence limit crossed the null (−7.4% [−15.0%, 0.2%]).

4. DISCUSSION

In this study of Mexican youth aged 8–14 years, *in utero* BPA and phthalate exposure was not associated with lipid profile during peripuberty. However, among boys, concurrent urinary levels of MCPP, MEP, and ΣDBP were each inversely related to total cholesterol and LDL-C; and MBzP was positively associated with HDL-C. In girls, higher concurrent urinary ΣDEHP concentrations corresponded with lower LDL-C.

Comparison of urinary EDC to other populations

In utero and peripubertal urinary EDC concentrations in the present analysis were similar to those reported in a previous publication that involved a subset of the participants (19) and an analysis of girls (26), both which also included a detailed comparison of analyte concentrations in the ELEMENT cohort to those of other populations (19). In brief, urinary BPA during the 3rd trimester, as well as the geometric mean of BPA across all of pregnancy, were comparable to those observed in Chinese adults (31) and pregnant Spanish women (32). Urinary concentrations of phthalates during pregnancy were also similar to those of U.S. adults, except for lower MBzP (geometric mean across pregnancy in ELEMENT: 245 ng/mL in ELEMENT vs. 959 ng/mL in adults 20 years in NHANES 2009–2010) (33). Additionally, we found similar urinary concentrations of phthalate metabolites to those reported for pregnant Israeli women during the 3rd trimester, except for MEP, which was higher in our study (geometric mean across pregnancy in ELEMENT: 235 ng/mL, ELEMENT 3rd trimester: 375 ng/mL, Israeli women 3rd trimester: 202 ng/mL) (34). In the children during peripuberty, urinary BPA and phthalate concentrations were similar to combined values reported for 6–11 year-old boys and girls in NHANES 2009–2010 (33), except for MBzP, which was lower in this study (ELEMENT boys: 7.4 ng/mL, ELEMENT girls: 9.5 ng/mL, U.S. boys and girls: 11.6 ng/mL).

Comparison of serum lipids during peripuberty to other populations

In comparison to a recent study of trends in serum lipids among children and adolescents 8–17 years in the U.S., prevalence of dyslipidemia in the ELEMENT population was lower: 6.1% vs. 7.8% with high total cholesterol, 4.0% vs. 12.8% with low HDL-C, and 1.6% vs. 8.4% with high LDL-C (1). Similarly, youth in our population had lower mean values for total cholesterol (155.2 vs. 160.3 mg/dL), triglycerides (87.5 vs. 57.8 mg/dL), and LDL (79.0 vs. 91.5 mg/dL), and higher HDL (58.7 vs. 52.2 mg/dL) as compared to 6–10 year old children in Project Viva, a Boston-area cohort (35). When we compared lipid profile of the ELEMENT population to that of adolescents attending public junior high schools in Mexico City (4), we observed comparable serum levels of total cholesterol, but higher HDL-C (59.3 vs. 43.9 mg/dL for boys, 57.6 vs. 44.9 mg/dL for girls) and lower LDL-C (75.9 vs. 91.6

mg/dL for boys, 81.9 vs. 96.3 mg/dL for girls) in both sexes, and lower triglycerides among boys only (77.0 vs. 87.7 mg/dL).

Associations of in utero and concurrent EDC exposure to lipid profile during peripuberty

Our finding that several phthalates were inversely associated with total cholesterol and LDL-C aligns with a longstanding animal literature. Three decades ago, Bell observed that dietary administration of DEHP inhibited cholesterolgenesis and decreased blood lipid levels in multiple animal species (7). More recent rodent models (8, 36) suggest that the hypolipidemic effect of DEHP may arise through its interactions with peroxisome proliferator-activated receptor- α (PPAR- α), a ligand-activated transcription factor that can decrease production of LDL-C and triglycerides, and upregulate HDL-C levels through its roles in lipid oxidation and fatty acid synthesis (37, 38). Likewise, our finding that higher peripubertal MEP was associated with lower total cholesterol and LDL-C could be explained by the fact that MEP is an estrogenic compound that may act as a PPAR- γ agonist (39) to reduce circulating lipid levels by promoting uptake and storage of free fatty acids in adipose tissue (40).

So far, investigations of phthalate exposure and circulating lipids in humans are scant and have focused on cross-sectional relationships of DEHP metabolites in rather specific adult populations. For example, higher serum MEHP, a phthalate metabolite indicative of ambient DEHP exposure, was associated with lower LDL-C in 1,016 elderly Swedes (10). Likewise, higher serum MEHP was associated with lower triglycerides, as well as lower levels of several fatty acid components, including palmitic, oleic, linoleic, and alpha-linoleic acids in 318 pregnant Japanese women (11). On the other hand, a recent study of 1,702 U.S. women in NHANES 1999–2004 found that urinary concentration of several DEHP metabolites was not associated with serum lipid profile (12). Yet, another analysis of 2,719 adults in NHANES 2001–2010, which aimed to explore relations of urinary phthalates with metabolic syndrome, revealed that higher MBzP and Σ DEHP corresponded greater odds of hypertriglyceridemia (serum triglycerides >150 mg/dL), particularly among women (13). The conflicting findings may be attributed, at least in part, to exposure misclassification as a result of use of different tissues to assess phthalate exposure – namely urine (12, 13) and serum (11) – although research in adults indicates moderate to strong correlations between phthalate concentrations in the two tissue types (i.e. R^2 of 0.45 to 0.90 for DEHP and DEP metabolites) (41–44), and inconsistent parameterization of the lipids as continuous (11, 12) vs. dichotomous outcomes (13). The limited, discrepant, and predominantly adult literature emphasizes the need for more studies examining associations of serum lipids with a broader range of phthalates in diverse populations.

Another explanation for our findings relates to tempo of maturation. During puberty, circulating levels of many lipids decrease (45, 46). Therefore, the inverse relations of MCP, MEP, and Σ DBP with total cholesterol and LDL-C in boys could be driven by pubertal status. In this population, we previously found that peripubertal urinary concentrations of Σ DBP metabolites MnBP and MiBP, and to a lesser extent MEP, correlated with more advanced puberty in boys based on testicular volume (47). Likewise, the inverse relationship between Σ DEHP and LDL-C in girls aligns with an earlier analysis in ELEMENT which

revealed a non-significant but consistent relations of peripubertal Σ DEHP metabolites MEHHP, MEOHP, and MECPP with greater odds of pubarche, thelarche, and menarche (26). Thus, although adjustment for pubertal status did not change our results, we cannot rule out the possibility of residual confounding by sexual maturation.

Of note, we did not observe any relationship between *in utero* EDC exposure and peripubertal lipid profile, which was unexpected in light of previous findings in this population that perinatal exposure to BPA and certain phthalates influenced several biomarkers of metabolic homeostasis during peripuberty (24). However, a recent rodent study found that *in utero* exposure to environmentally-relevant levels of BPA resulted in higher fat mass at birth, as well as altered expression of several genes involved in hepatic lipid metabolism at the time of weaning (which roughly corresponds with the age of our study participants), but no differences in circulating lipids (48). It is possible that EDCs induce upstream physiological alterations that may not manifest in circulating biomarkers until later in the life course – a prospect that deserves additional investigation as the ELEMENT youth transition into adulthood.

Strengths of this study include our ability to examine the potential effects of BPA and phthalate exposure during two sensitive periods for metabolic programming on peripubertal lipid profile, and our ability to account for physician-assessed pubertal status and evaluate for differences by sex. Limitations of this study include the relatively small sample size, the cross-sectional peripubertal analysis which precludes our ability to infer on temporality or causality, and potential exposure misclassification.

5. Conclusions

In summary, while *in utero* EDC exposure was not related to lipid profile during peripuberty, we observed sex-specific associations with respect to concurrent phthalate exposure. Specifically, in boys, higher urinary levels of MCP, MEP, and Σ DBP at 8–14 years was related to lower total cholesterol and LDL-C, while MBzP was associated with higher HDL-C. In girls, higher urinary Σ DEHP correlated with lower LDL-C. Although the effect sizes we observed were relatively small (i.e., differences of –11.3 to –8.6 mg/dL for total cholesterol, –9.6 to –7.5 mg/dL for LDL, 6.6 mg/dL for HDL among boys; difference of –6.5 mg/dL of LDL in girls), they may be relevant in the long term given that even a 1 mg/dL increment in LDL at 11 years of age was found to predict nearly two times higher odds of metabolic syndrome in young adulthood among Bogalusa Heart Study participants (49). Nevertheless, considering that adolescence is a time of rapid physiological change and high within-person variability in metabolic parameters – including circulating lipid levels (50) – additional studies are warranted to investigate whether our findings that concurrent exposure to certain phthalates are associated with a more favorable lipid profile persists beyond adolescence.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

We thank the American British Cowdry Hospital for providing us with research facilities.

FUNDING: This work was supported by the following grants: P01ES022844 and T32ES007062 from the National Institute for Environmental Health Sciences (NIEHS), and RD83543601 from the US Environmental Protection Agency (US EPA). This study was also supported and partially funded by the National Institute of Public Health/Ministry of Health of Mexico. It contents are solely the responsibility of the grantee and do not necessarily represent the official views of the US EPA. Further, the US EPA does not endorse the purchase any commercial products or services mentioned in the publication.

Abbreviations

BPA	Bisphenol A
DBP	dibutyl phthalate metabolites
DEHP	di-2-ethylhexyl phthalate metabolites
LOD	limit of detection
HDL-C	high density lipoprotein cholesterol
LDL-C	low density lipoprotein cholesterol
MBzP	monobenzyl phthalate
MCPP	mono-3-carboxypropyl phthalate
MECPP	mono-2-ethyl-5-carboxypentyl phthalate
MEHHP	mono-2-ethyl-5-hydroxyhexyl phthalate
MEHP	mono-2-ethylhexyl phthalate
MEP	monoethyl phthalate
MEOHP	mono-2-ethyl-5-oxohexyl phthalate
MiBP	mono-isobutyl phthalate
MnBP	mono- <i>n</i> -butyl phthalate

References

1. Kit BK, Kuklina E, Carroll MD, Ostchega Y, Freedman DS, Ogden CL. Prevalence of and trends in dyslipidemia and blood pressure among us children and adolescents, 1999–2012. *JAMA Pediatrics*. 2015; 169(3):272–9. [PubMed: 25599372]
2. American Academy of Pediatrics. Expert panel on integrated guidelines for cardiovascular health and risk reduction in children and adolescents: summary report. *Pediatrics*. 2011; 128(Suppl 5):S213–56. [PubMed: 22084329]
3. Yamamoto-Kimura L, Posadas-Romero C, Posadas-Sanchez R, Zamora-Gonzalez J, Cardoso-Saldana G, Mendez Ramirez I. Prevalence and interrelations of cardiovascular risk factors in urban and rural Mexican adolescents. *The Journal of adolescent health : official publication of the Society for Adolescent Medicine*. 2006; 38(5):591–8. [PubMed: 16635772]

4. Posadas-Sanchez R, Posadas-Romero C, Zamora-Gonzalez J, Mendoza-Perez E, Cardoso-Saldana G, Yamamoto-Kimura L. Lipid and lipoprotein profiles and prevalence of dyslipidemia in Mexican adolescents. *Metabolism: clinical and experimental*. 2007; 56(12):1666–72. [PubMed: 17998019]
5. Juonala M, Viikari JS, Ronnema T, Marniemi J, Jula A, Loo BM, et al. Associations of dyslipidemias from childhood to adulthood with carotid intima-media thickness, elasticity, and brachial flow-mediated dilatation in adulthood: the Cardiovascular Risk in Young Finns Study. *Arteriosclerosis, thrombosis, and vascular biology*. 2008; 28(5):1012–7.
6. Grun F, Blumberg B. Endocrine disruptors as obesogens. *Molecular and cellular endocrinology*. 2009; 304(1–2):19–29. [PubMed: 19433244]
7. Bell FP. Effects of phthalate esters on lipid metabolism in various tissues, cells and organelles in mammals. *Environmental health perspectives*. 1982; 45:41–50. [PubMed: 7140695]
8. Hayashi Y, Ito Y, Yamagishi N, Yanagiba Y, Tamada H, Wang D, et al. Hepatic peroxisome proliferator-activated receptor alpha may have an important role in the toxic effects of di(2-ethylhexyl)phthalate on offspring of mice. *Toxicology*. 2011; 289(1):1–10. [PubMed: 21354252]
9. Moghaddam HS, Samarghandian S, Farkhondeh T. Effect of bisphenol A on blood glucose, lipid profile and oxidative stress indices in adult male mice. *Toxicology mechanisms and methods*. 2015; 25(7):507–13. [PubMed: 26376105]
10. Olsen L, Lind L, Lind PM. Associations between circulating levels of bisphenol A and phthalate metabolites and coronary risk in the elderly. *Ecotoxicology and environmental safety*. 2012; 80:179–83. [PubMed: 22421452]
11. Jia X, Harada Y, Tagawa M, Naito H, Hayashi Y, Yetti H, et al. Prenatal maternal blood triglyceride and fatty acid levels in relation to exposure to di(2-ethylhexyl)phthalate: a cross-sectional study. *Environmental Health and Preventive Medicine*. 2014; 20(3):168–78. [PubMed: 25543208]
12. Yaghjian L, Sites S, Ruan Y, Chang SH. Associations of urinary phthalates with body mass index, waist circumference and serum lipids among females: National Health and Nutrition Examination Survey 1999–2004. *International journal of obesity (2005)*. 2015; 39(6):994–1000. [PubMed: 25644057]
13. James-Todd TM, Huang T, Seely EW, Saxena AR. The association between phthalates and metabolic syndrome: the National Health and Nutrition Examination Survey 2001–2010. *Environmental health : a global access science source*. 2016; 15:52. [PubMed: 27079661]
14. Webber LS, Srinivasan SR, Wattigney WA, Berenson GS. Tracking of serum lipids and lipoproteins from childhood to adulthood. The Bogalusa Heart Study. *American journal of epidemiology*. 1991; 133(9):884–99. [PubMed: 2028978]
15. Sadler TW. Susceptible periods during embryogenesis of the heart and endocrine glands. *Environmental health perspectives*. 2000; 108(Suppl 3):555–61. [PubMed: 10852854]
16. Selevan SG, Kimmel CA, Mendola P. Identifying critical windows of exposure for children's health. *Environmental health perspectives*. 2000; 108(Suppl 3):451–5. [PubMed: 10852844]
17. Hoyt LT, Falconi A. Puberty and Perimenopause: Reproductive Transitions and their Implications for Women's Health. *Social science & medicine (1982)*. 2015; 132:103–12. [PubMed: 25797100]
18. Dietz WH. Critical periods in childhood for the development of obesity. *The American journal of clinical nutrition*. 1994; 59(5):955–9. [PubMed: 8172099]
19. Lewis RC, Meeker JD, Peterson KE, Lee JM, Pace GG, Cantoral A, et al. Predictors of urinary bisphenol A and phthalate metabolite concentrations in Mexican children. *Chemosphere*. 2013; 93(10):2390–8. [PubMed: 24041567]
20. Rivera JA, de Cossio TG, Pedraza LS, Aburto TC, Sanchez TG, Martorell R. Childhood and adolescent overweight and obesity in Latin America: a systematic review. *The lancet Diabetes & endocrinology*. 2014; 2(4):321–32. [PubMed: 24703050]
21. OECD. Obesity Update. 2014
22. Food and agriculture organization (FAO) of the United Nations. Rome: 2013.
23. Hu H, Tellez-Rojo MM, Bellinger D, Smith D, Ettinger AS, Lamadrid-Figueroa H, et al. Fetal lead exposure at each stage of pregnancy as a predictor of infant mental development. *Environmental health perspectives*. 2006; 114(11):1730–5. [PubMed: 17107860]

24. Watkins DJ, Peterson KE, Ferguson KK, Mercado-Garcia A, Ortiz MT, Cantoral A, et al. Relating phthalate and BPA exposure to metabolism in peripubescence: The role of exposure timing, sex, and puberty. *The Journal of clinical endocrinology and metabolism*. 2015;jc20152706.
25. Silva MJ, Samandar E, Preau JL Jr, Reidy JA, Needham LL, Calafat AM. Quantification of 22 phthalate metabolites in human urine. *Journal of chromatography B, Analytical technologies in the biomedical and life sciences*. 2007; 860(1):106–12. [PubMed: 17997365]
26. Watkins DJ, Tellez-Rojo MM, Ferguson KK, Lee JM, Solano-Gonzalez M, Blank-Goldenberg C, et al. In utero and peripubertal exposure to phthalates and BPA in relation to female sexual maturation. *Environmental research*. 2014; 134:233–41. [PubMed: 25173057]
27. Ferguson KK, McElrath TF, Ko Y-A, Mukherjee B, Meeker JD. Variability in urinary phthalate metabolite levels across pregnancy and sensitive windows of exposure for the risk of preterm birth. *Environment international*. 2014; 70:118–24. [PubMed: 24934852]
28. Chen Y-H, Ferguson KK, Meeker JD, McElrath TF, Mukherjee B. Statistical methods for modeling repeated measures of maternal environmental exposure biomarkers during pregnancy in association with preterm birth. *Environmental Health*. 2015; 14:9. [PubMed: 25619201]
29. de Onis M, Onyango AW, Borghi E, Siyam A, Nishida C, Siekmann J. Development of a WHO growth reference for school-aged children and adolescents. *Bull World Health Organ*. 2007; 85(9): 660–7. [PubMed: 18026621]
30. U.S. Department of Health and Human Services. Plan and operation of the Third National Health and Nutrition Examination Survey, 1988–94. Series 1: programs and collection procedures. *Vital and health statistics Ser 1, Programs and collection procedures*. 1994; (32):1–407.
31. He Y, Miao M, Herrinton LJ, Wu C, Yuan W, Zhou Z, et al. Bisphenol A levels in blood and urine in a Chinese population and the personal factors affecting the levels. *Environmental research*. 2009; 109(5):629–33. [PubMed: 19426969]
32. Casas M, Valvi D, Luque N, Ballesteros-Gomez A, Carsin AE, Fernandez MF, et al. Dietary and sociodemographic determinants of bisphenol A urine concentrations in pregnant women and children. *Environment international*. 2013; 56:10–8. [PubMed: 23542682]
33. Centers for Disease Control and Prevention (CDC). Fourth national report on human exposure to environmental chemicals. 2013
34. Berman T, Hochner-Celnikier D, Calafat AM, Needham LL, Amitai Y, Wormser U, et al. Phthalate exposure among pregnant women in Jerusalem, Israel: Results of a pilot study. *Environment international*. 2009; 35(2):353–7. [PubMed: 18824263]
35. Perng W, Hajj H, Belfort MB, Rifas-Shiman SL, Kramer MS, Gillman MW, et al. Birth Size, Early Life Weight Gain, and Midchildhood Cardiometabolic Health. *The Journal of pediatrics*. 2016; 173:122–30.e1. [PubMed: 26995700]
36. Nakashima R, Hayashi Y, Md K, Jia X, Wang D, Naito H, et al. Exposure to DEHP decreased four fatty acid levels in plasma of prepartum mice. *Toxicology*. 2013; 309:52–60. [PubMed: 23619606]
37. Kimura R, Takahashi N, Murota K, Yamada Y, Niiya S, Kanzaki N, et al. Activation of peroxisome proliferator-activated receptor- α (PPAR α) suppresses postprandial lipidemia through fatty acid oxidation in enterocytes. *Biochemical and biophysical research communications*. 2011; 410(1):1–6. [PubMed: 21640707]
38. Colin S, Briand O, Touche V, Wouters K, Baron M, Pattou F, et al. Activation of intestinal peroxisome proliferator-activated receptor- α increases high-density lipoprotein production. *European heart journal*. 2013; 34(32):2566–74. [PubMed: 22843443]
39. Guven C, Dal F, Aydogan Ahbab M, Taskin E, Ahbab S, Adin Cinar S, et al. Low dose monoethyl phthalate (MEP) exposure triggers proliferation by activating PDX-1 at 1.1B4 human pancreatic beta cells. *Food and chemical toxicology : an international journal published for the British Industrial Biological Research Association*. 2016; 93:41–50. [PubMed: 27133914]
40. Laplante M, Festuccia WT, Soucy G, Gelinas Y, Lalonde J, Deshaies Y. Involvement of adipose tissues in the early hypolipidemic action of PPAR γ agonism in the rat. *American journal of physiology Regulatory, integrative and comparative physiology*. 2007; 292(4):R1408–17.
41. Kato K, Silva MJ, Reidy JA, Hurtz D 3rd, Malek NA, Needham LL, et al. Mono(2-ethyl-5-hydroxyhexyl) phthalate and mono-(2-ethyl-5-oxohexyl) phthalate as biomarkers for human

- exposure assessment to di-(2-ethylhexyl) phthalate. *Environmental health perspectives*. 2004; 112(3):327–30. [PubMed: 14998748]
42. Hogberg J, Hanberg A, Berglund M, Skerfving S, Remberger M, Calafat AM, et al. Phthalate diesters and their metabolites in human breast milk, blood or serum, and urine as biomarkers of exposure in vulnerable populations. *Environmental health perspectives*. 2008; 116(3):334–9. [PubMed: 18335100]
43. Hines EP, Calafat AM, Silva MJ, Mendola P, Fenton SE. Concentrations of phthalate metabolites in milk, urine, saliva, and Serum of lactating North Carolina women. *Environmental health perspectives*. 2009; 117(1):86–92. [PubMed: 19165392]
44. Frederiksen H, Jorgensen N, Andersson AM. Correlations between phthalate metabolites in urine, serum, and seminal plasma from young Danish men determined by isotope dilution liquid chromatography tandem mass spectrometry. *Journal of analytical toxicology*. 2010; 34(7):400–10. [PubMed: 20822678]
45. Mascarenhas LP, Leite N, Titski AC, Brito LM, Boguszewski MC. Variability of lipid and lipoprotein concentrations during puberty in Brazilian boys. *Journal of pediatric endocrinology & metabolism : JPEM*. 2015; 28(1–2):125–31. [PubMed: 25153571]
46. Eissa MA, Mihalopoulos NL, Holubkov R, Dai S, Labarthe DR. Changes in Fasting Lipids during Puberty. *The Journal of pediatrics*. 2016; 170:199–205. [PubMed: 26706233]
47. Ferguson KK, Peterson KE, Lee JM, Mercado-Garcia A, Blank-Goldenberg C, Tellez-Rojo MM, et al. Prenatal and peripubertal phthalates and bisphenol A in relation to sex hormones and puberty in boys. *Reproductive toxicology* (Elmsford, NY). 2014; 47:70–6.
48. Somm E, Schwitzgebel VM, Toulotte A, Cederroth CR, Combescure C, Nef S, et al. Perinatal exposure to bisphenol A alters early adipogenesis in the rat. *Environmental health perspectives*. 2009; 117(10):1549–55. [PubMed: 20019905]
49. Camhi SM, Katzmarzyk PT, Broyles S, Srinivasan SR, Chen W, Bouchard C, et al. Predicting adult body mass index-specific metabolic risk from childhood. *Metabolic syndrome and related disorders*. 2010; 8(2):165–72. [PubMed: 20156071]
50. Haney, EM., Huffman, LH., Bougatsos, Cea. *Screening for Lipid Disorders in Children and Adolescents*. Rockville (MD): Agency for Healthcare Research and Quality (US); 2007. Available from: <http://www.ncbi.nlm.nih.gov/books/NBK33489/>

HUMAN SUBJECTS IRB

The institutional review boards of the Mexico National Institute of Public Health and the University of Michigan approved research protocols. Maternal informed consent and child assent were provided for all participants.

Distribution of serum lipid levels at age 8–14 years across background characteristics of 248 ELEMENT participants.

Table 1

	N	Total cholesterol (mg/dL)		HDL-C (mg/dL)		LDL-C (mg/dL)		Triglycerides (mg/dL)	
		Mean ± SD	p ^a	Mean ± SD	p ^a	Mean ± SD	p ^a	Mean ± SD	p ^a
Overall	248	155.2 ± 28.4		58.7 ± 11.9		79.0 ± 23.2		87.5 ± 44.4	
Sociodemographic & family characteristics									
Maternal age at enrollment									
15–24 years	101	153.3 ± 30.6	0.34	57.9 ± 12.2	0.47	76.9 ± 24.2	0.23	92.6 ± 47.9	0.51
25–34 years	116	156.2 ± 25.8		59.5 ± 12.4		80.3 ± 21.9		81.7 ± 39.9	
35–44 years	31	158.2 ± 30.4		58.2 ± 9.3		81.5 ± 24.5		92.7 ± 47.4	
Marital status									
Married	177	155.7 ± 30.4	0.64	59.4 ± 11.9	0.12	79.3 ± 24.5	0.75	85.0 ± 43.8	0.11
Single	71	154.1 ± 22.8		56.9 ± 12.0		78.4 ± 19.6		93.8 ± 45.6	
Maternal education									
<10 years	89	153.3 ± 29.0	0.10	57.9 ± 12.5	0.58	77.1 ± 23.5	0.08	91.5 ± 47.9	0.98
10–12 years	123	154.0 ± 27.2		59.5 ± 12.2		78.3 ± 22.5		81.0 ± 34.5	
>12 years	36	164.1 ± 30.0		57.8 ± 9.5		86.3 ± 23.8		100.2 ± 60.4	
Characteristics at the 8–14 year research visit									
Sex									
Male	117	151.2 ± 28.5	0.03	59.9 ± 12.0	0.11	75.9 ± 23.9	0.04	77.0 ± 38.3	0.0001
Female	131	158.8 ± 27.8		57.6 ± 11.8		81.9 ± 22.2		96.9 ± 47.4	
BMI z-score									
< -2.0	4	164.5 ± 16.2	0.003	68.3 ± 8.3	0.0001	81.2 ± 15.0	0.0004	75.5 ± 38.3	<0.0001
-2.0 to 1.0	119	148.9 ± 25.2		61.5 ± 12.4		72.6 ± 21.1		73.8 ± 36.0	
>1.0 to 2.0	83	159.7 ± 28.9		55.8 ± 10.7		84.6 ± 22.2		96.6 ± 45.0	
>2.0	42	163.5 ± 33.2		55.5 ± 11.3		86.1 ± 26.8		109.9 ± 52.5	
Pubertal status: Males ^b									
Prepubertal	56	155.6 ± 26.9	0.17	60.9 ± 11.7	0.35	79.4 ± 21.5	0.20	76.4 ± 39.7	0.80
Pubertal	58	148.2 ± 29.3		59.0 ± 12.3		73.7 ± 25.6		77.8 ± 38.2	
Pubertal status: Females ^b									
Prepubertal	86	165.2 ± 26.6	0.0002	59.1 ± 11.7	0.04	87.1 ± 21.0	0.0002	95.3 ± 49.1	0.34

Author Manuscript

Author Manuscript

Author Manuscript

Author Manuscript

	N	Total cholesterol (mg/dL)		HDL-C (mg/dL)		LDL-C (mg/dL)		Triglycerides (mg/dL)	
		Mean ± SD	<i>p</i> ^a	Mean ± SD	<i>p</i> ^a	Mean ± SD	<i>p</i> ^a	Mean ± SD	<i>p</i> ^a
Pubertal	45	146.6 ± 26.3		54.6 ± 11.6		72.0 ± 21.3		100.1 ± 44.3	

^aRepresents a test for linear trend where an ordinal indicator is entered into the model as continuous variable, with the exception of binary variables (Wald test).

^bPrepubertal and pubertal were defined as Tanner stage 1 and >1, respectively, for genital or pubic hair development in boys, and for breast or pubic hair development in girls.

Table 2

Associations of geometric mean maternal urinary BPA and phthalate concentrations across pregnancy (“*in utero*” exposure) with offspring serum lipid levels at 8–14 years among ELEMENT mother-child pairs ($n = 227$).

Maternal urinary concentrations ^c	Mean $\pm$ SD ^a	% Difference (95% CI) in serum lipid levels during peripuberty per IQR in maternal urinary BPA and phthalate concentrations ^b	LDL-C	HDL-C ^c	Triglycerides ^c
BPA	1.1 $\pm$ 1.0 ng/mL	-2.0 (-5.7, 1.8)	-4.0 (-10.0, 2.0)	-1.0 (-5.2, 3.4)	0.4 (-8.8, 10.6)
MBzP	4.7 $\pm$ 4.7 ng/mL	-1.8 (-5.2, 1.7)	-2.4 (-7.9, 3.2)	-0.9 (-4.8, 3.1)	-2.7 (-10.9, 6.3)
MCPP	1.5 $\pm$ 1.2 ng/mL	0.7 (-3.2, 4.7)	1.4 (-5.0, 7.9)	-1.1 (-5.6, 3.6)	2.7 (-7.3, 13.7)
MEP	245 $\pm$ 370 ng/mL	-1.5 (-5.4, 2.4)	0 (-6.3, 6.3)	-4.0 (-8.3, 0.4)	0.8 (-8.8, 11.5)
Σ DEHP	0.3 $\pm$ 0.3 nmol/mL	-1.2 (-4.9, 2.4)	-2.1 (-8.0, 3.8)	-2.0 (-6.1, 2.3)	2.6 (-6.6, 12.7)
Σ DBP	0.4 $\pm$ 0.4 nmol/mL	0.1 (-3.6, 3.7)	1.4 (-4.5, 7.3)	-1.7 (-5.8, 2.5)	0.5 (-8.5, 10.4)

^aValues are not corrected for dilution.

^bEstimates are adjusted for child's age at the time of lipid assessment, sex, and urinary specific gravity.

^cNatural-log (ln) transformed for analysis

Sex-specific associations of concurrent urinary concentrations of BPA and phthalate metabolites with serum lipid levels at 8–14 years among ELEMENT youth.

Table 3

Concurrent urinary concentrations ^c	Mean ± SD ^a	% Difference (95% CI) in serum lipids during peripuberty per IQR increment in urinary BPA and phthalate concentrations ^b			
		Total Cholesterol	LDL-C	HDL-C ^e	Triglycerides ^c
Boys (<i>n</i> = 112)					
BPA	2.0 ± 4.2 ng/mL	-0.7 (-5.8, 4.3)	-4.6 (-12.9, 3.8)	3.4 (-2.4, 9.5)	-0.7 (-12.9, 13.2)
MBzP	7.4 ± 5.9 ng/mL	-0.7 (-6.3, 4.4)	-7.2 (-16.3, 2.0)	6.6 (0.2, 13.5)	0.8 (-12.8, 16.4)
MCPP	2.7 ± 2.6 ng/mL	-7.4 (-12.8, -2.0)	-12.7 (-21.6, -3.8)	-0.8 (-6.9, 5.7)	-7.2 (-19.6, 7.1)
MEP	173 ± 354 ng/mL	-5.7 (-10.4, -1.0)	-10.8 (-18.5, -3.1)	1.0 (-4.4, 6.7)	-2.9 (-14.2, 10.0)
ΣDEHP	1.7 ± 11.0 nmol/mL	-0.5 (-4.6, 3.7)	-1.3 (-8.1, 5.5)	2.1 (-2.6, 6.9)	-5.3 (-14.8, 5.3)
ΣDBP	0.7 ± 0.7 nmol/mL	-6.7 (-11.8, -1.6)	-9.9 (-18.4, -1.5)	-5.2 (-10.6, 0.6)	-1.9 (-14.4, 12.3)
Girls (<i>n</i> = 128)					
BPA	2.0 ± 2.6 ng/mL	0.1 (-4.5, 4.7)	-0.2 (-7.4, 7.1)	-1.9 (-7.3, 3.8)	8.8 (-3.5, 22.6)
MBzP	9.5 ± 16.7 ng/mL	-0.8 (-5.4, 3.9)	-0.5 (-7.9, 6.9)	-1.0 (-6.6, 4.9)	2.4 (-9.4, 15.8)
MCPP	4.7 ± 13.1 ng/mL	-1.4 (-5.2, 2.4)	-4.3 (-10.2, 1.7)	1.2 (-3.5, 6.0)	2.1 (-7.6, 12.9)
MEP	335 ± 797 ng/mL	-1.7 (-5.5, 2.2)	4.5 (-10.5, 1.5)	1.4 (-3.3, 6.4)	0.8 (-8.9, 11.6)
ΣDEHP	0.6 ± 0.6 nmol/mL	-4.3 (-9.1, 0.5)	-7.9 (-15.4, -0.4)	-3.5 (-9.1, 2.4)	4.5 (-8.0, 18.7)
ΣDBP	1.1 ± 2.8 nmol/mL	-1.4 (-5.4, 2.7)	-5.9 (-12.3, 0.5)	4.3 (-0.8, 9.7)	1.0 (-9.3, 12.4)

^aValues are not corrected for dilution.

^bEstimates are adjusted for child's age at the time of lipid assessment and urinary specific gravity.

^cNatural-log (ln) transformed.