

Original Article

The Association of Six Phthalate Metabolites with Body Mass Index and Waist Circumference in NHANES 1999-2002

Elizabeth E. Hatch¹, Jessica W. Nelson², M. Mustafa Qureshi³, Janice Weinberg⁴, Lynn L. Moore³, Martha Singer³, Thomas F. Webster²

¹ Department of Epidemiology, Boston University School of Public Health

² Department of Environmental Health, Boston University School of Public Health

³ Department of Preventive Medicine, Boston University School of Medicine

⁴ Department of Biostatistics, Boston University School of Public Health

Corresponding author:

Elizabeth E. Hatch

Associate Professor of Epidemiology

Boston University School of Public Health

715 Albany St, Talbot 318 East

Boston, MA 02118

617-638-7791 (phone)

617-638-4458 (fax)

eehatch@bu.edu

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Abstract

Background: Although diet and activity are key factors in the obesity epidemic, laboratory studies suggest that endocrine disrupting chemicals may also affect obesity.

Methods: We analyzed associations between six phthalate metabolites measured in urine and body mass index (BMI) and waist circumference (WC) in National Health and Nutrition Examination Survey (NHANES) participants aged 6-80. We included 4369 participants from NHANES 1999-2002, with data on mono-ethyl (MEP), mono-2-ethylhexyl (MEHP), mono-n-butyl (MBP), and mono-benzyl (MBzP) phthalate; 2286 also had data on mono-2-ethyl-5-hydroxyhexyl (MEHHP) and mono-2-ethyl-5-oxohexyl (MEOHP) phthalate (2001-2002). Using multiple regression, we computed mean BMI and WC within phthalate quartiles in eight age/gender specific models.

Results: The most consistent associations were in males aged 20-59; BMI and WC increased across quartiles of MBzP (adjusted mean BMI=26.7, 27.2, 28.4, 29.0, p-trend=0.0002), and positive associations were also found for MEOHP, MEHHP, MEP, and MBP. In females, BMI and WC increased with MEP quartile in adolescent girls (adjusted mean BMI=22.9, 23.8, 24.1, 24.7, p-trend =0.03), and a similar but less strong pattern was seen in 20-59 year olds. In contrast, MEHP was inversely related to BMI in adolescent girls (adjusted mean BMI=25.4, 23.8, 23.4, 22.9, p-trend=0.02) and females aged 20-59 (adjusted mean BMI=29.9, 29.9, 27.9, 27.6, p-trend=0.02). There were no important associations among children, but several inverse associations among 60-80 year olds.

Conclusions: This exploratory, cross-sectional analysis revealed a number of interesting associations with different phthalate metabolites and obesity outcomes, including notable differences by gender and age subgroups. Effects of endocrine disruptors, such as phthalates, may depend upon endogenous hormone levels, which vary dramatically by age and gender. Individual phthalates also have different biologic and hormonal effects. Although our study has limitations, both of these factors could explain some of the variation in the observed associations. These preliminary data support the need for prospective studies in populations at risk for obesity.

Keywords= phthalates, endocrine disruptors, obesity, obesogens, BMI, waist circumference, NHANES

Introduction

The prevalence of obesity has increased dramatically over the last several decades in the United States (U.S.) and Europe, and more recently in developing countries [1]. Although changes in dietary patterns and physical activity are undoubtedly key causal factors [2], there is growing interest in the possibility that endocrine disrupting chemicals, such as phthalates, may affect obesity-related pathways by altering hormone action or gene expression [3, 4]. Exposure to endocrine disrupting chemicals can interfere with synthesis, release, transport, metabolism, binding, action, or elimination of natural hormones. Proposed biologic mechanisms that could underlie an association between endocrine disruptors and obesity include alterations in thyroid and steroid hormone function [5, 6], and activation of peroxisome proliferator-activated receptors (PPARs) [7] which play a major role in adipocyte differentiation and energy storage [8].

Phthalates are ubiquitous industrial chemicals used as plasticizers, solvents, lubricants, and stabilizers in the manufacture of consumer products such as children's toys, medical equipment and medications, cosmetics, and food packaging [9]. The general population is exposed to phthalates by ingestion of food and water, dermal exposure, and inhalation of polluted air [10]. Once taken into the body, phthalates are quickly metabolized and excreted in urine and feces. Lower molecular weight phthalates, such as diethyl phthalate (DEP), are metabolized primarily to their hydrolytic monoesters during phase I biotransformation, whereas higher molecular weight phthalates, including di-2-ethylhexyl phthalate (DEHP), are further metabolized to more hydrophilic oxidative metabolites. Monoesters and oxidative metabolites can be excreted unchanged or can

undergo glucuronidation in a phase II biotransformation to a more water soluble compound [9, 10].

Animal studies have found carcinogenic effects, testicular and ovarian toxicity, and hormonal, hepatic, and renal effects following phthalate exposure [9, 11-14]. While the results of human studies are not entirely consistent [9], several have suggested anti-androgenic effects [15-17] and one reported thyroid function changes [18], both of which may play a role in obesity and fat distribution. The only study to directly evaluate phthalates and obesity-related outcomes in humans found a positive correlation between several phthalate metabolites and waist circumference and a measure of insulin resistance among adult males[19].

The U.S. Centers for Disease Control and Prevention (CDC) has conducted biomonitoring on random samples of approximately one-third of the total National Health and Nutrition Examination Survey (NHANES) population since 1999. Over 100 environmental chemicals, including phthalate metabolites, have been measured in blood and/or urine, and are publicly available for the years 1999-2002 [10]. In this analysis, we examined the relationship between six urinary phthalate metabolites and body mass index (BMI) and waist circumference (WC) in male and female subjects, aged 6-80 years, who were participants in NHANES during 1999-2002. Because both body size and hormonal processes vary substantially with age and gender, we conducted stratified analyses within eight age/gender subgroups (6-11, 12-19, 20-59, and 60-80 years).

Methods

NHANES is a nationally representative random sample of the non-institutionalized U.S. civilian population selected using a complex multistage probability sampling design. Since 1999, NHANES has enrolled approximately 5000 subjects each year. In-home interviews and physical examinations in a mobile exam unit are used to assess behavioral and physical risk factors and characteristics. NHANES obtained written informed consent from all participants, and all data are available on the NHANES website in a de-identified form. Details regarding questionnaires, physical examination components, and testing procedures are available elsewhere [20].

A total of 5149 participants, aged 6 and older, had phthalate measurements and data on BMI and WC. We excluded participants who reported dialysis (n=4), chemotherapy (n=8), insulin treatment (n=64), pregnancy (n=204), or breastfeeding (n=39) because of associations with BMI or central adiposity. We also excluded 148 subjects over 80 years old as weight loss commonly occurs in the elderly. Finally, we excluded 313 subjects who had missing values on one or more covariates.

Measurement of phthalate metabolites

Details of laboratory methods for measuring phthalate metabolites are reported elsewhere [21]. Briefly, spot urine samples were frozen at -20°C and shipped to CDC's National Center for Environmental Health for analysis. Metabolites were measured using isotope dilution high-performance liquid chromatography and mass spectrometry. NHANES measured seven phthalate mono-ester metabolites in urine samples during 1999-2002: mono-ethyl phthalate (MEP), mono-(2-ethyl)-hexyl phthalate (MEHP),

mono-benzyl phthalate (MBzP), mono-cyclohexyl phthalate (MCP), mono-isononyl phthalate (MNP), mono-n-octyl phthalate (MOP), and mono-butyl phthalate (MBP), which represents the sum of two isomers, mono-n-butyl and mono-isobutyl phthalate. We analyzed four metabolites detectable in at least 80% of the study population: MEP, MEHP, MBP, and MBzP. Several additional metabolites were measured during 2001-2002. We included mono-(2-ethyl-5-hydroxyhexyl) phthalate (MEHHP) and mono-(2-ethyl-5-oxohexyl) phthalate (MEOHP), two oxidative metabolites of MEHP that are detected at higher levels than MEHP [22, 23]. Metabolites less than the limit of detection (LOD) were replaced with the LOD divided by the square root of two [10]. A total of 4369 observations were available for the analyses of MEP, MEHP, MBP, and MBzP, and 2286 subjects for the analyses of MEHHP and MEOHP.

Anthropometric variables

NHANES measured weight, height, and WC during the physical examination according to a standard protocol [24]. We used BMI as a measure of overall obesity and WC as an indicator of central adiposity, which may be more strongly related to health than BMI [25].

Diet, exercise, and other potential confounding variables

Dietary variables: NHANES assessed diet with a 24-hour recall using a computer-assisted dietary interview. We examined the following dietary variables as potential confounders of the relation between phthalates and obesity/central obesity: (1) energy intake; (2) energy-adjusted macronutrient intakes (total fat, saturated fat,

carbohydrates, and protein); (3) grams of fiber per 1000 kilocalories; (4) daily servings in each of the five major U.S. Department of Agriculture (USDA) Food Pyramid categories [26] (dairy, fruit, grains, vegetables, and meat); and (5) discretionary fat and added sugars as defined by the USDA Food Pyramid.

Physical activity: Physical activity was assessed for subjects aged 12 and older with a standard leisure time activity questionnaire. Estimated energy expenditure was derived by applying metabolic equivalent (MET) levels to reported frequencies of activities. We also investigated confounding effects of physical activity using other variables, including a dichotomous variable for leisure time physical activity; average minutes/day of leisure activity; a variable which asked respondents whether their activity was greater, the same, or less than others their age; and an index which combined several leisure and non-leisure time activity variables.

We estimated sedentary behavior by hours spent watching television, playing video games, and using a computer outside of work. Parents provided information on their children's behavior during the past 24-hour time period; adolescents (aged 12-19) and adults reported their usual daily habits over the past month.

Demographic variables: NHANES collected data on race/ethnicity, education, and family income during the home interview. Education level was categorized as less than high school, high school diploma, and more than high school. We used three alternative approaches to examine potential confounding effects of socio-economic status (SES): self-reported total family income; the poverty-income ratio (PIR, a ratio of U.S. Census-defined poverty level to family income); and a dichotomous indicator which combined several socioeconomic variables. The indicator variable first used income, if

available, to categorize participants as lower SES if their income was less than or equal to \$20,000. If data on income were missing, education was used to classify participants (aged 19 and older) as low SES if they had less than high school education. If both income and education were missing, evidence of food insecurity among parents or children (e.g., use of food stamps, insufficient food for parents or children) was used to classify subjects as low SES.

Smoking and alcohol consumption: Information on smoking and alcohol consumption was available for adults aged 20 and older. Based on information reported in the home interview, we classified subjects as current, former, or never smokers. We categorized alcohol consumption habits (during the prior month) as none, < 1 drink per week, 1-<4 drinks per week, 4-<8 drinks per week, or 8 or more drinks per week.

Menopausal status and parity: We used questions on menstrual history to categorize women aged 20 and older as pre- or postmenopausal. Women who were still having regular menstrual cycles (not due to hormone replacement therapy) were considered premenopausal. We classified women as postmenopausal if they had not had a menstrual period for ≥ 12 months or had had a hysterectomy. Women whose status was unclear were assigned missing values. Parity was categorized into nulliparous, one live birth, or more than one live birth.

Statistical analysis

We carried out gender specific analyses within four age strata: 6-11, 12-19, 20-59, and 60-80 years. We used a locally weighted regression smoother (LOESS) [27] to examine the shape of the relationship between each phthalate as a continuous variable

and BMI or WC. As these analyses did not identify natural cut-points, we used age/gender specific quartiles of each metabolite as the exposure variables in multivariate models. We also created quartiles based on the overall distribution of each phthalate in the entire population aged 6-80.

We used multiple linear regression models to examine the relationship between phthalate quartiles and BMI and WC controlling for potential confounding variables. We included age, creatinine, race/ethnicity, height, and SES as core covariates in all models. Age was modeled continuously in children aged 6-11 and adolescents aged 12-19, and categorically (by 10 year age group) in adults. As recommended by Barr et al, we included creatinine as an independent variable in the model, instead of using creatinine-adjusted phthalate values [28]. We included race/ethnicity and SES because of relationships with both phthalates [10, 29] and BMI and WC [30, 31]. As there were no important differences between the three SES variables described above, we selected the dichotomous variable for the final models because it had the fewest missing values. Additional potential confounders were retained if their inclusion led to meaningful changes in the exposure effect estimates in any of the age and gender subgroups. Variables included in the final models were percent of calories from total fat (<30, 30-38, 38+), servings of dairy (tertiles), servings of fruits and vegetables (tertiles), METS/month, TV/video/computer use, smoking (aged 20+), menopausal status, and parity (females aged 20+). Neither alcohol consumption nor education had meaningful effects on the phthalate estimates. Tests for trend were performed by treating phthalate category as a linear predictor in the models.

In evaluating the relationship between MEHP and the outcomes, we ran models that included MEHP's oxidative metabolites separately as covariates in addition to the standard model described above. Hauser [32] and Meeker [18] used this method to address potential individual differences in metabolism of DEHP.

SAS version 9.1 was used for all analyses. We used Proc Surveyreg to calculate adjusted mean differences in BMI and WC accounting for stratification by geography and the proportion of minority populations and clustering within primary sampling units. We adjusted for the covariates representing the over-sampled subgroups rather than using sampling weights; this adjustment is regarded as a good compromise between efficiency and bias [33, 34].

Results

Table 1 presents summary statistics for measures of obesity, exposure, and covariates by age and gender. Phthalates were detected in most participants; MEP and MBP were detected in greater than 99%, MBzP and MEHHP in 98%, MEOHP in 97%, and MEHP in 81% of the study sample. MEP was detected at the highest levels in all subgroups. Females had higher levels of all metabolites than males. Except for MEP, levels of phthalates were highest in 6-11 year olds. Females had higher mean BMI in all age groups except children, whereas males had higher mean WC measurements. The high proportion of Hispanic and non-Hispanic black adolescents is attributable to over-sampling in that age group.

Phthalate metabolites were moderately or strongly correlated (Table 2). MEP had the weakest correlations ($r < 0.26-0.38$) with other phthalates. MEHP was strongly

correlated with the oxidative metabolites of DEHP, MEHHP and MEOHP ($r=0.68$), which were highly correlated with each other ($r=0.98$). MBP and MBzP were also strongly correlated ($r=0.72$).

The associations between phthalate metabolites and the outcomes, BMI and WC, were similar; therefore, we only present the results for BMI graphically. Results for both BMI and WC are included in Appendix Tables 1 and 2. Figures 1-5 present adjusted mean differences in BMI (kg/m^2) with increasing quartile of phthalate exposure for five metabolites, separately for males and females. Results of trend tests are included for trends with $p\text{-values}<0.15$. The results for MEHHP and MEOHP were virtually identical (MEOHP not shown). We found no major differences in the results when we used cut-points based on quartiles derived from the entire study population; therefore, we present the results for age/gender specific quartiles. Appendix Tables 1 and 2 present the ranges for each phthalate quartile, within age and gender subgroups, and the regression coefficients for both BMI (kg/m^2) and WC (cm) comparing quartiles 2-4 to quartile 1 separately by age, along with 95% confidence limits (CIs) and the $p\text{-values}$ for test of trend.

Figures 1A and B present results for MEP for males and females respectively. A positive relationship between MEP quartile and BMI was apparent for adult males (20-59 and 60-80), and for adolescent and adult (20-59) females. Most of the coefficients for MEP were positive, with the exception of adolescent males (no relationship) and older females (an inverse relationship). For adolescent girls, adjusted mean BMIs were 22.9, 23.8, 24.1, and 24.7 ($p\text{-trend}=0.03$) with increasing quartile of metabolite concentration, and adjusted mean WCs were 77.4, 79.7, 80.1, and 81.6 ($p\text{-trend}=0.02$). The effect

estimate for WC for the highest quartile of MEP was larger than all other variables in the fully adjusted model; the next largest estimates were -3.1 and -2.9 cm for the top two tertiles of fruit and vegetable consumption, compared to 4.1 and 2.7 cm for the highest two MEP quartiles compared to the lowest. The associations between MEP and BMI and WC in adult women were similar, but less pronounced; the adjusted mean BMIs among women aged 20-59 by MEP quartiles were 28.9, 28.9, 29.0, and 29.8 (p-trend=0.14). There was some evidence for further effect modification by age with positive effects for 40-59 year old females, but little evidence of an association among 20-39 year olds.

Figures 2A and B present results for MBP. There were significant inverse trends between MBP level and BMI and WC among 60-80 year olds. There were no clear trends among children or adolescents of either gender. The patterns in adult males and females were different, with suggestive positive trends among males for both BMI and WC, but inverse trends among females.

There was a strong positive relationship between MBzP quartile and BMI and WC among adult males aged 20-59 (Figure 3A). The adjusted mean BMIs from quartile 1 to 4 were 26.7, 27.2, 28.4, and 29.0 (p-trend=0.0002). In contrast, there was a borderline inverse trend among 60-80 year old males. There were no major trends among females, although the estimates were positive among all age groups, except for 60-80 year olds.

MEHP was inversely related to BMI and WC in adolescent and adult females (Figure 4B). Mean BMI generally decreased across quartiles among adolescent girls (25.1, 23.7, 23.0, and 23.6 from the lowest to highest quartiles of MEHP, p-trend=0.02) and among 20-59 year old females (29.8, 30.2, 28.5, and 28.1, p-trend=0.02). Results

were similar among 60-80 year old females. There were no major trends between MEHP and either BMI or WC among males, except for a borderline inverse trend among 60-80 year old males. Results for MEHP were not affected by including either of the oxidative metabolites in the model (data not shown).

Figures 5A and B show the results for MEHHP, one of the oxidative metabolites of MEHP. There was a positive relationship between MEHHP and BMI among 20-59 year old males, and a non-monotonic increase among 12-19 year old males (Figure 5A). For MEHHP, BMI increased from 27.1 to 28.8 between quartiles 1 and 4 (p-trend=0.10) and WC increased from 95.2 to 99.8 (p-trend=0.08) among 20-59 year old males. There were no important trends in BMI or WC among females; for 60-80 year olds the estimates were inverse, whereas they were positive for children and adolescents.

Discussion

This exploratory, cross-sectional analysis of six phthalate metabolites revealed a number of interesting associations with BMI and WC, including several dose-response relationships. Once considered to be a passive organ for energy storage, adipose tissue is now known to be an active endocrine organ that secretes numerous chemical signals and responds to a variety of hormonal signals [35, 36] and thus may plausibly be affected by endocrine disrupting chemicals. Animal evidence and limited human data suggest that phthalates have the potential to affect obesity through several different biologic mechanisms that may differ by phthalate (Table 3). These include anti-androgenic effects, inhibition of thyroid hormone action, and activation of PPARs [7, 37]. PPAR γ in particular has been shown to play a major role in adipocyte differentiation and energy

storage [8]; its activation promotes the differentiation of preadipose cells into adipocytes [37].

Our results showed a striking difference in effect between males and females. The strongest positive effects were seen among 20-59 males for MBzP, MEHHP, and MEOHP. In contrast, findings among females varied by metabolite, with some positive effects for MEP, particularly among adolescents, but inverse associations for MEHP and MBzP. Variation in the effect of phthalates by gender is plausible. Several phthalates, as mentioned, are anti-androgens; lower free testosterone levels have been found in males exposed to higher levels of DBP and DEHP [16]. Higher testosterone is associated with smaller WC and a better cardiovascular profile in males, whereas higher androgen levels in females are associated with higher BMI, greater risk for metabolic syndrome, and conditions such as polycystic ovarian disease [38]. Therefore, women with the highest levels of MEHP may have lower levels of androgens or a higher estrogen/androgen ratio, which could explain the inverse relationship between MEHP and BMI. Our results appear somewhat consistent with an anti-androgenic effect of certain phthalates, although additional explanations may be involved, particularly for MEP.

In addition to the gender differences, the associations also differed by age group. We found no associations between phthalates and body size among 6-11 year olds, even though this group had the highest levels of all phthalate metabolites except MEP. In part, this may be due to the smaller sample sizes in this subgroup. BMI and WC are also highly variable in children by age and development status, related in part to timing of the adiposity rebound [39], which could result in less ability to detect effects of external factors. Because metabolite levels are higher in children (except for MEP), the lowest

quartile of exposure calculated by age group does not necessarily represent the same low exposure as in other age groups. Even so, results among the 6-11 year olds were substantially the same when exposure quartiles were based on the overall distribution of phthalates in the total study population compared to the results based on the age/gender specific quartiles.

We found mixed associations among adolescents and 20-59 year olds. Different effects by age are plausible because physiology and endogenous hormone levels vary by age in both males and females, potentially modifying the effects of endocrine disruptors. The observed associations among 60-80 year olds were quite different. In general, BMI and WC declined with increasing levels of phthalate metabolites. These results are surprising and deserve further exploration. It is also possible that a physiological change in older people is related to both weight loss and altered phthalate metabolism.

Only one prior study has evaluated the association between phthalates and obesity in humans. Stahlhut [19] also used NHANES data to evaluate the relationship between phthalates and WC, along with a measure of insulin resistance, in adult males. The authors reported similar, mostly positive associations between waist circumference and several phthalates in adult males. Results for children, adolescents, and females were not reported. There were some differences in analytic approach; for example, Stahlhut evaluated all males aged 18 and older, whereas we evaluated 20-59 year olds and 60-80 year olds separately after noticing distinctly different associations in the two groups. We used quartiles of exposure after determining that there were no clear patterns from smoothed regression models, whereas Stahlhut used continuous, log transformed phthalate values, in addition to quintiles of exposure. There were also differences in the

confounders that were evaluated in the two studies; in particular, we evaluated a larger number of dietary variables in our models. The qualitative similarity of our results for adult males with those of Stahlhut et al [34] suggest they are reasonably robust to differences in analysis methods. The striking differences between males and females revealed by our analysis significantly add to the earlier work and appears consistent with endocrine disruption by certain phthalates.

Strengths of our study include the broad range of exposures, relatively good statistical power even within subgroups, and detailed evaluation of available potential confounding variables. The ability to evaluate effect modification by age and gender was critical as we saw striking differences in results by these variables. We tested multiple dietary factors, four different measures of physical activity, and three alternative measures of SES. We also evaluated the data using exposure quartiles based on the entire study population in addition to age/gender derived quartiles of exposure, and found that the results were not materially changed.

Our study has several important limitations. Its cross-sectional nature precludes the ability to make any causal inferences about the direction of the association between phthalates and obesity. Although phthalates are rapidly metabolized and do not accumulate in adipose tissue, it is unknown if heavier individuals metabolize phthalates differently than do individuals with less body fat. Elevated exposures to phthalates have been reported due to use of certain medications [40], and it is possible that some of our results may be due to greater use of medications among heavier individuals. It is also possible that our results, particularly for MEP, may be due to greater use of lotions and cosmetics in people with a larger body surface area, resulting in a higher internal and

excreted dose. However, due to the multiple environmental sources of DEP, this seems unlikely.

We controlled for numerous confounding variables, including diet and physical activity. However, diet was measured using a 24-hour recall, which has limitations for estimating long-term dietary patterns, and physical activity was measured over the 30 days prior to the interview, which may not reflect long-term patterns. Although NHANES collects data on a wide variety of potential confounders, there may be other unknown or unmeasured confounders. Some of our findings may be due to chance, as we evaluated associations between six phthalate metabolites and two outcomes in eight population sub-groups.

Exposure to mixtures of endocrine disrupting chemicals may have different effects than single exposures [41]. We were unable to examine combined exposures to multiple types of chemicals because NHANES measured chemicals in different population samples. While this dataset does include measurements for multiple phthalates, it is not clear how these exposures should be combined when looking at the relationship between phthalates and body size. Compounds may have different and even potentially opposite effects, and multiple mechanisms may be involved. While work is being done to look at cumulative effects of phthalate exposure on reproductive tract development [42], there currently exist insufficient animal and human data to develop a good biologically-based exposure index for potential effects on obesity.

Single spot-urine measurements of phthalates were used to estimate exposure. Phthalates are rapidly metabolized and excreted, and a single exposure measurement may not reflect long-term exposure. Exposure sources may vary over time based on dietary

intake, use of personal care products, medications, and other factors. Several studies have evaluated the consistency of phthalate measurements over time and, in general, low to moderate correlation coefficients were found [43-45], although moderate to high sensitivity to classify individuals into the highest tertile of exposure has been reported [44]. Therefore, our effect estimates are likely affected by non-differential misclassification and comparisons between highest and lowest quartiles may be biased towards the null.

A final limitation of the study is that, due to its cross-sectional nature, we could not evaluate exposures throughout the life course, particularly the sensitive window of prenatal and neonatal development. In utero and early life exposures may be critical; they may permanently alter gene expression patterns that affect metabolic processes [4].

Our results are exploratory and based on cross-sectional data, but suggest that further research on the potential for phthalates to act as obesogens is warranted. We observed several associations that were not only statistically significant but of magnitudes of clinical relevance. Prospective studies in human populations are needed to evaluate prenatal and neonatal exposures, and to look at exposure to multiple endocrine disrupting chemicals.

References

1. Ebbeling, C.B., D.B. Pawlak, and D.S. Ludwig, *Childhood obesity: public-health crisis, common sense cure*. Lancet, 2002. **360**(9331): p. 473-82.
2. Hill, J.O. and J.C. Peters, *Environmental contributions to the obesity epidemic*. Science, 1998. **280**(5368): p. 1371-4.
3. Grun, F. and B. Blumberg, *Environmental obesogens: organotins and endocrine disruption via nuclear receptor signaling*. Endocrinology, 2006. **147**(6 Suppl): p. S50-5.
4. Heindel, J.J., *Endocrine disruptors and the obesity epidemic*. Toxicol Sci, 2003. **76**(2): p. 247-9.
5. Knudsen, N., et al., *Small differences in thyroid function may be important for body mass index and the occurrence of obesity in the population*. J Clin Endocrinol Metab, 2005. **90**(7): p. 4019-24.
6. Mayes, J.S. and G.H. Watson, *Direct effects of sex steroid hormones on adipose tissues and obesity*. Obes Rev, 2004. **5**(4): p. 197-216.
7. Hurst, C.H. and D.J. Waxman, *Activation of PPARalpha and PPARgamma by environmental phthalate monoesters*. Toxicol Sci, 2003. **74**(2): p. 297-308.
8. Barak, Y. and S. Kim, *Genetic Manipulations of PPARs: Effects on Obesity and Metabolic Disease*. PPAR Res, 2007. **2007**: p. 12781.
9. Hauser, R. and A.M. Calafat, *Phthalates and human health*. Occup Environ Med, 2005. **62**(11): p. 806-18.
10. Centers for Disease Control and Prevention (CDC), N.C.f.H.S.N., *Third National Report on Human Exposure to Environmental Chemicals*. 2005, CDC: Atlanta.
11. ATSDR, *Toxicological profile for Di-n-butyl phthalate (DBP)*. . 2001, Agency for Toxic Substances and Disease Registry, Atlanta GA.
12. ATSDR, *Toxicological Profile for Di (2-ethylhexyl) phthalate (DEHP)*. . 2002, Agency for Toxic Substances and Disease Registry, Atlanta, GA.
13. Davis, B.J., R.R. Maronpot, and J.J. Heindel, *Di-(2-ethylhexyl) phthalate suppresses estradiol and ovulation in cycling rats*. Toxicol Appl Pharmacol, 1994. **128**(2): p. 216-23.
14. Gray, L.E., Jr., et al., *Perinatal exposure to the phthalates DEHP, BBP, and DINP, but not DEP, DMP, or DOTP, alters sexual differentiation of the male rat*. Toxicol Sci, 2000. **58**(2): p. 350-65.
15. Main, K.M., et al., *Human breast milk contamination with phthalates and alterations of endogenous reproductive hormones in infants three months of age*. Environ Health Perspect, 2006. **114**(2): p. 270-6.
16. Pan, G., et al., *Decreased serum free testosterone in workers exposed to high levels of di-n-butyl phthalate (DBP) and di-2-ethylhexyl phthalate (DEHP): a cross-sectional study in China*. Environ Health Perspect, 2006. **114**(11): p. 1643-8.

17. Swan, S.H., et al., *Decrease in anogenital distance among male infants with prenatal phthalate exposure*. Environ Health Perspect, 2005. **113**(8): p. 1056-61.
18. Meeker, J.D., A.M. Calafat, and R. Hauser, *Di-(2-ethylhexyl) Phthalate Metabolites May Alter Thyroid Hormone Levels in Men*. Environ Health Perspect, 2007. **115**: p. 1029-1034.
19. Stahlhut, R.W., et al., *Concentrations of Urinary Phthalate Metabolites are Associated with Increased Waist Circumference and Insulin Resistance in Adult U.S. Males*. Environ Health Perspect, 2007. **115**: p. 876-882.
20. Centers for Disease Control and Prevention (CDC), N.C.f.H.S.N. 1999-2002 [cited; Available from:
<http://www.cdc.gov/nchs/about/major/nhanes/datalink.htm>.
21. Silva, M.J., et al., *Urinary levels of seven phthalate metabolites in the U.S. population from the National Health and Nutrition Examination Survey (NHANES) 1999-2000*. Environ Health Perspect, 2004. **112**(3): p. 331-8.
22. Barr, D.B., et al., *Assessing human exposure to phthalates using monoesters and their oxidized metabolites as biomarkers*. Environ Health Perspect, 2003. **111**(9): p. 1148-51.
23. Weuve, J., et al., *Exposure to phthalates in neonatal intensive care unit infants: urinary concentrations of monoesters and oxidative metabolites*. Environ Health Perspect, 2006. **114**(9): p. 1424-31.
24. Centers for Disease Control and Prevention (CDC), N.C.f.H.S.N., *Anthropometry Procedures Manual*. 2002.
25. Janssen, I., P.T. Katzmarzyk, and R. Ross, *Waist circumference and not body mass index explains obesity-related health risk*. Am J Clin Nutr, 2004. **79**(3): p. 379-84.
26. (USDA), U.S.D.o.A., *The Food Guide Pyramid*. 1992.
27. Hastie TJ, T.R., *Generalized Additive Models*. 1990, Chapman and Hall: London. p. 29-31.
28. Barr, D.B., et al., *Urinary creatinine concentrations in the U.S. population: implications for urinary biologic monitoring measurements*. Environ Health Perspect, 2005. **113**(2): p. 192-200.
29. Koo, J.W., et al., *The association between biomarker-based exposure estimates for phthalates and demographic factors in a human reference population*. Environ Health Perspect, 2002. **110**(4): p. 405-10.
30. Ogden, C.L., et al., *Prevalence of Overweight and Obesity in the United States, 1999-2004*. JAMA, 2006. **295**(13): p. 1549-1555.
31. Zhang, Q. and Y. Wang, *Socioeconomic inequality of obesity in the United States: do gender, age, and ethnicity matter?* Social Science & Medicine, 2004. **58**(6): p. 1171-1180.
32. Hauser, R., et al., *DNA damage in human sperm is related to urinary levels of phthalate monoester and oxidative metabolites*. Hum Reprod, 2007. **22**(3): p. 688-95.
33. Graubard, B.I. and E.L. Korn, *Analyzing health surveys for cancer-related objectives*. J Natl Cancer Inst, 1999. **91**(12): p. 1005-16.
34. Korn, E.L. and B.I. Graubard, *Epidemiologic studies utilizing surveys: accounting for the sampling design*. Am J Public Health, 1991. **81**(9): p. 1166-73.

35. Ahima, R.S. and J.S. Flier, *Adipose tissue as an endocrine organ*. Trends Endocrinol Metab, 2000. **11**(8): p. 327-32.
36. Cooke, P.S. and A. Naaz, *Role of estrogens in adipocyte development and function*. Exp Biol Med (Maywood), 2004. **229**(11): p. 1127-35.
37. Feige, J.N.G., L. Rossi, D., Zoete, V., Metivier, R., Tudor, C., Anghel, S.I., Grosdidier, A., Lathion, C., Engelborghs, Y., Michielin, O., Wahli, W., Desvergne, B., *The endocrine disruptor mono-ethyl-hexyl-phthalate is a selective PPARgamma modulator which promotes adipogenesis*. J. Biol Chem.
38. Barber, T.M., et al., *Obesity and polycystic ovary syndrome*. Clinical Endocrinology, 2006. **65**(2): p. 137-145.
39. Rolland-Cachera, M.F., et al., *Early adiposity rebound: causes and consequences for obesity in children and adults*. Int J Obes. **30**(S4): p. S11-S17.
40. Hauser, R., et al., *Medications as a source of human exposure to phthalates*. Environ Health Perspect, 2004. **112**(6): p. 751-3.
41. Kortenkamp, A., *Ten Years of Mixing Cocktails – a Review of Combination Effects of Endocrine Disrupting Chemicals*. . Environmental Health Perspectives 2007 **115**(Supplement 1): p. 98-105.
42. Howdeshell, K.L., et al., *Cumulative effects of dibutyl phthalate and diethylhexyl phthalate on male rat reproductive tract development: altered fetal steroid hormones and genes*. Toxicol Sci, 2007. **99**(1): p. 190-202.
43. Fromme, H., et al., *Occurrence and daily variation of phthalate metabolites in the urine of an adult population*. Int J Hyg Environ Health, 2007. **210**(1): p. 21-33.
44. Hauser, R., et al., *Temporal variability of urinary phthalate metabolite levels in men of reproductive age*. Environ Health Perspect, 2004. **112**(17): p. 1734-40.
45. Hoppin, J.A., et al., *Reproducibility of urinary phthalate metabolites in first morning urine samples*. Environ Health Perspect, 2002. **110**(5): p. 515-8.
46. Bility, M.T., et al., *Activation of mouse and human peroxisome proliferator-activated receptors (PPARs) by phthalate monoesters*. Toxicol Sci, 2004. **82**(1): p. 170-82.
47. Ishihara, A., et al., *The effect of endocrine disrupting chemicals on thyroid hormone binding to Japanese quail transthyretin and thyroid hormone receptor*. Gen Comp Endocrinol, 2003. **134**(1): p. 36-43.
48. Sugiyama, S., et al., *Detection of thyroid system-disrupting chemicals using in vitro and in vivo screening assays in Xenopus laevis*. Toxicol Sci, 2005. **88**(2): p. 367-74.
49. Breous, E., A. Wenzel, and U. Loos, *The promoter of the human sodium/iodide symporter responds to certain phthalate plasticisers*. Mol Cell Endocrinol, 2005. **244**(1-2): p. 75-8.
50. Shimada, N. and K. Yamauchi, *Characteristics of 3,5,3'-triiodothyronine (T3)-uptake system of tadpole red blood cells: effect of endocrine-disrupting chemicals on cellular T3 response*. J Endocrinol, 2004. **183**(3): p. 627-37.
51. Wenzel, A., et al., *Modulation of iodide uptake by dialkyl phthalate plasticisers in FRTL-5 rat thyroid follicular cells*. Mol Cell Endocrinol, 2005. **244**(1-2): p. 63-71.

52. Fisher, J.S., *Environmental anti-androgens and male reproductive health: focus on phthalates and testicular dysgenesis syndrome*. *Reproduction*, 2004. **127**(3): p. 305-15.
53. Gray, L.E., Jr., et al., *Adverse effects of environmental antiandrogens and androgens on reproductive development in mammals*. *Int J Androl*, 2006. **29**(1): p. 96-104; discussion 105-8.
54. Lampen, A., S. Zimnik, and H. Nau, *Teratogenic phthalate esters and metabolites activate the nuclear receptors PPARs and induce differentiation of F9 cells*. *Toxicol Appl Pharmacol*, 2003. **188**(1): p. 14-23.
55. Maloney, E.K. and D.J. Waxman, *trans-Activation of PPARalpha and PPARgamma by structurally diverse environmental chemicals*. *Toxicol Appl Pharmacol*, 1999. **161**(2): p. 209-18.

Figure Titles and Legends

Figure 1: Difference in Body Mass Index, by Increasing MEP Quartile and Age Group. (A) displays results for males, (B) for females. All estimates are adjusted for age, race/ethnicity, creatinine, height, SES, dietary factors, TV, Mets/month (age 12+), smoking (age 20+), and reproductive factors (age 20+ females). Results of trend tests are included for those with p-values<0.15.

Figure 2: Difference in Body Mass Index, by Increasing MBP Quartile and Age Group. (A) displays results for males, (B) for females. All estimates are adjusted for age, race/ethnicity, creatinine, height, SES, dietary factors, TV, Mets/month (age 12+), smoking (age 20+), and reproductive factors (age 20+ females). Results of trend tests are included for those with p-values<0.15.

Figure 3: Difference in Body Mass Index, by Increasing MBzP Quartile and Age Group. (A) displays results for males, (B) for females. All estimates are adjusted for age, race/ethnicity, creatinine, height, SES, dietary factors, TV, Mets/month (age 12+), smoking (age 20+), and reproductive factors (age 20+ females). Results of trend tests are included for those with p-values<0.15.

Figure 4: Difference in Body Mass Index, by Increasing MEHP Quartile and Age Group. (A) displays results for males, (B) for females. All estimates are adjusted for age, race/ethnicity, creatinine, height, SES, dietary factors, TV, Mets/month (age 12+), smoking (age 20+), and reproductive factors (age 20+ females). Results of trend tests are included for those with p-values<0.15.

Figure 5: Difference in Body Mass Index, by Increasing MEHHP Quartile and Age Group. (A) displays results for males, (B) for females. All estimates are adjusted for age, race/ethnicity, creatinine, height, SES, dietary factors, TV, Mets/month (age 12+), smoking (age 20+), and reproductive factors (age 20+ females). Results of trend tests are included for those with p-values<0.15.

Table 1. Distribution of selected characteristics by age and gender subgroup, NHANES 1999-2002, phthalate subsample

Characteristics	Females				Males			
	6-11 yrs (n=327)	12-19 yrs (n=682)	20-59 yrs (n=761)	60-80 yrs (n=348)	6-11 yrs (n=329)	12-19 yrs (n=662)	20-59 yrs (n=895)	60-80 yrs (n=365)
Phthalate metabolites ¹ (% above LOD ²)	<i>Geometric mean (SD)</i>							
MEP (99)	137.7 (2.8)	207.3 (3.2)	225.6 (3.3)	218.6 (3.5)	93.9 (2.9)	137.0 (3.5)	182.1 (4.1)	184.8 (5.5)
MBP (99)	48.0 (2.3)	30.8 (2.3)	24.6 (2.3)	25.7 (2.4)	38.0 (2.2)	19.3 (2.2)	15.3 (2.1)	17.5 (2.5)
MBzP (98)	34.4 (2.8)	17.1 (2.7)	12.8 (2.7)	11.7 (2.7)	34.7 (2.6)	15.7 (2.8)	10.1 (2.5)	9.5 (3.1)
MEHP (81)	5.4 (2.8)	3.8 (2.9)	4.0 (2.9)	3.3 (2.9)	5.5 (3.1)	2.7 (3.0)	3.3 (3.2)	2.5 (2.9)
MEOHP (97)	27.5 (2.4)	15.0 (2.4)	12.5 (2.7)	12.4 (2.6)	26.6 (2.4)	12.2 (2.8)	10.6 (2.8)	9.2 (2.7)
MEHHP (98)	39.6 (2.5)	21.1 (2.6)	18.3 (2.8)	18.4 (2.7)	39.1 (2.4)	18.2 (2.8)	16.6 (3.0)	13.2 (2.9)
	<i>Arithmetic mean (SD)</i>							
Age (yrs)	8.6 (1.7)	15.2 (2.3)	39.5 (11.0)	68.4 (5.94)	8.6 (1.6)	15.4 (2.30)	38.8 (11.1)	68.7 (6.0)
BMI (kg/m ²)	18.5 (3.9)	24.0 (5.7)	28.9 (7.3)	29.2 (6.2)	18.7 (4.1)	23.6 (5.5)	27.9 (5.6)	28.1 (4.7)
Waist circumference (cm)	64.2 (11.1)	80.2 (13.3)	92.8 (15.3)	97.5 (14.1)	65.3 (12.1)	81.8 (15.1)	97.6 (15.3)	103.6 (12.1)
Height (cm)	135.3 (13.8)	160.9 (7.2)	161.8 (6.8)	158.6 (6.8)	135.6 (11.5)	169.3 (11.1)	175.5 (7.9)	172.2 (7.3)
Creatinine (mg/dL)	109.1 (55.5)	162.0 (87.0)	124.9 (79.5)	92.5 (60.4)	110.2 (57.2)	176.0 (93.7)	166.8 (91.1)	132.7 (77.0)
Physical activity (met equiv hrs/month)	N/A	165.4 (331.5)	57.1 (118.1)	40.2 (92.7)	N/A	217.8 (295.4)	96.4 (196.1)	59.6 (141.3)
Dietary intake								
% kilocalories from total fat	33.1 (7.5)	31.9 (8.8)	33.5 (10.0)	32.1 (9.5)	33.2 (7.3)	32.1 (7.8)	31.8 (10.0)	33.1 (9.6)
Fruit & veg (servings/day)	3.5 (2.5)	4.0 (3.3)	4.1 (2.9)	4.78 (3.2)	3.8 (2.8)	4.1 (3.9)	5.1 (4.2)	5.0 (3.9)
Dairy (servings/day)	1.8 (1.3)	1.5 (1.4)	1.3 (1.4)	1.2 (1.4)	2.1 (1.4)	2.0 (1.8)	1.6 (1.9)	1.4 (1.6)
	<i>Column percent</i>							
Socioeconomic status (% low)	33.9	35.5	26.7	44.5	35.0	35.5	27.3	35.1
Race/ethnicity								
African American	33.3	30.9	19.8	18.7	33.7	29.5	19.7	16.7
Hispanic	33.9	39.6	30.0	27.0	35.0	40.0	29.8	26.6
Whites and others	32.7	29.5	50.2	54.3	31.3	30.5	50.5	56.7
Television viewing (% >2.5 hrs/day)	48.9	51.6	36.4	52.9	53.2	56.2	43.9	48.0

Smoking (% current)	N/A	N/A	23.3	12.9	N/A	N/A	31.1	16.4
¹ $\mu\text{g/g}$ creatinine								
² level of detection								

Table 2. Spearman Correlation Coefficients* between phthalate metabolites, NHANES 1999-2002

	MEP	MBP	MBzP	MEHP	MEHHP	MEOHP
MEP	1	0.38	0.34	0.26	0.32	0.33
MBP		1	0.72	0.46	0.62	0.64
MBzP			1	0.39	0.58	0.59
MEHP				1	0.68	0.68
MEHHP					1	0.98
MEOHP						1

* All coefficients are significant at $p < 0.001$

Table 3: Biological Effects of Phthalates Potentially Related to Obesity

	PPAR-γ activation	Thyroid effect	Anti-androgenic effect
DEP/MEP	- [7, 46]	- [47, 48]	+/- [9]
DBP/MBP	+/- [7]	+/- [47-51]	++ [9, 16, 52, 53]
BBzP/MBzP	+ [7, 37]	++ [47-51]	++ [9, 52, 53]
DEHP/MEHP	++ [7, 37, 46, 54, 55]	+/- [18, 47-51]	+ [9, 16, 52, 53]

++ strong, consistent effects
 + moderate, consistent effects
 +/- inconsistent effects
 - no effect

Appendix Table 1. Quartile ranges for 6 phthalate metabolites, and adjusted* mean difference in BMI (kg/m^2) and WC (cm) by quartile, among females by age group, NHANES 1999-2002

MEP	N	Range (mg/dL)	BMI β (95% CI)	p-trend	WC β (95% CI)	p-trend
Age 6-11						
Quartile 4	82	307.0 - 1836.9	0.30 (-1.22, 1.81)	0.66	1.05 (-3.30, 5.40)	0.61
Quartile 3	82	129.8 - 305.6	0.54 (-0.58, 1.67)		0.99 (-2.05, 4.02)	
Quartile 2	82	54.8 - 129.6	0.35 (-0.86, 1.56)		0.74 (-2.64, 4.12)	
Quartile 1	81	0.6 - 54.4	Referent		Referent	
Age 12-19						
Quartile 4	170	694.5 - 39631.7	1.74 (-0.02, 3.49)	0.03	4.11 (0.37, 7.86)	0.02
Quartile 3	171	265.9 - 694.3	1.16 (-0.43, 2.75)		2.7 (-0.83, 6.23)	
Quartile 2	171	110.8 - 265.7	0.84 (-0.59, 2.26)		2.31 (-0.80, 5.42)	
Quartile 1	170	5.9 - 110.6	Referent		Referent	
Age 20-59						
Quartile 4	190	512.9 - 21932.2	0.92 (-0.51, 2.36)	0.14	2.07 (-0.72, 4.85)	0.1
Quartile 3	191	219.5 - 512.0	0.10 (-1.02, 1.22)		0.46 (-2.16, 3.07)	
Quartile 2	190	91.0 - 217.7	-0.03 (-1.33, 1.26)		0.07 (-2.67, 2.80)	
Quartile 1	190	1.7 - 90.5	Referent		Referent	
Age 60-80						
Quartile 4	86	424.0 - 14346.1	-0.21 (-1.73, 1.32)	0.64	-0.22 (-3.49, 3.04)	0.82
Quartile 3	86	143.2 - 396.4	-0.79 (-2.24, 0.66)		-1.62 (-4.99, 1.76)	
Quartile 2	86	56.3 - 142.0	-0.03 (-1.91, 1.85)		-0.62 (-4.29, 3.06)	
Quartile 1	86	3.4 - 55.6	Referent		Referent	
MBP	N	Range (mg/dL)	BMI β (95% CI)	p-trend	WC β (95% CI)	p-trend
Age 6-11						
Quartile 4	82	92.2 - 2595.3	0.07 (-1.12, 1.27)	0.55	0.37 (-2.67, 3.40)	0.84
Quartile 3	82	51.0 - 92.0	0.43 (-0.90, 1.77)		0.69 (-2.74, 4.12)	
Quartile 2	82	24.7 - 50.8	0.35 (-0.75, 1.45)		0.63 (-2.39, 3.64)	
Quartile 1	81	0.6 - 24.5	Referent		Referent	
Age 12-19						
Quartile 4	172	84.1 - 1192.9	-0.17 (-2.24, 1.90)	0.2	-0.47 (-4.71, 3.77)	0.31
Quartile 3	169	46.5 - 83.7	0.17 (-1.60, 1.94)		0.38 (-3.46, 4.23)	
Quartile 2	171	22.3 - 46.3	0.37 (-1.20, 1.93)		1.08 (-2.05, 4.22)	
Quartile 1	170	0.6 - 22.1	Referent		Referent	
Age 20-59						
Quartile 4	191	51.7 - 4693.3	-1.43 (-3.37, 0.52)	0.29	-2.60 (-6.15, 0.95)	0.24
Quartile 3	189	26.7 - 51.6	0.04 (-1.82, 1.90)		-0.06 (-3.33, 3.21)	
Quartile 2	191	11.7 - 26.7	-0.68 (-1.78, 0.41)		-0.61 (-2.87, 1.65)	
Quartile 1	190	0.6 - 11.6	Referent		Referent	

Age 60-80

Quartile 4	87	42.0 - 639.1	-2.69 (-4.54, -0.84)	0.01	-5.67 (-9.31, -2.03)	0.02
Quartile 3	87	19.0 - 41.7	-1.26 (-2.70, 0.18)		-3.94 (-7.47, -0.41)	
Quartile 2	87	9.3 - 18.9	-0.87 (-2.70, 0.96)		-1.85 (-6.19, 2.50)	
Quartile 1	87	0.6 - 9.3	Referent		Referent	

MBzP	N	Range (mg/dL)	BMI β (95% CI)	p-trend	WC β (95% CI)	p-trend
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Age 6-11

Quartile 4	81	68.8 - 1685.0	-0.18 (-1.43, 1.08)	0.80	-0.50 (-3.66, 2.66)	0.65
Quartile 3	83	35.7 - 68.3	0.68 (-0.50, 1.85)		1.33 (-1.75, 4.41)	
Quartile 2	81	14.5 - 34.2	0.52 (-0.89, 1.92)		1.69 (-1.63, 5.02)	
Quartile 1	82	0.6 - 14.4	Referent		Referent	

Age 12-19

Quartile 4	170	56.9 - 739.7	0.84 (-0.97, 2.65)	0.59	1.46 (-3.06, 5.98)	0.74
Quartile 3	171	26.9 - 55.9	0.53 (-0.96, 2.01)		0.59 (-2.86, 4.05)	
Quartile 2	171	11.1 - 26.4	1.42 (-0.07, 2.91)		2.48 (-0.68, 5.64)	
Quartile 1	170	0.2 - 11.0	Referent		Referent	

Age 20-59

Quartile 4	190	29.8 - 1009.4	0.82 (-1.26, 2.90)	0.62	3.18 (-0.90, 7.26)	0.29
Quartile 3	190	14.6 - 29.7	0.78 (-1.01, 2.56)		2.08 (-1.62, 5.79)	
Quartile 2	192	5.8 - 14.5	1.26 (-0.11, 2.62)		3.55 (0.51, 6.59)	
Quartile 1	189	0.2 - 5.6	Referent		Referent	

Age 60-80

Quartile 4	87	21.8 - 331.9	-0.73 (-2.67, 1.22)	0.49	-2.41 (-6.65, 1.84)	0.24
Quartile 3	88	9.1 - 21.6	-0.72 (-2.85, 1.40)		-2.18 (-6.26, 1.91)	
Quartile 2	84	3.8 - 9.0	-0.67 (-2.13, 0.80)		-1.33 (-5.24, 2.59)	
Quartile 1	89	0.2 - 3.7	Referent		Referent	

MEHP	N	Range (mg/dL)	BMI β (95% CI)	p-trend	WC β (95% CI)	p-trend
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Age 6-11

Quartile 4	82	11.1 - 314.3	-0.90 (-2.51, 0.71)	0.45	-2.51 (-6.52, 1.49)	0.33
Quartile 3	82	5.2 - 10.9	0.17 (-1.24, 1.59)		-0.37 (-3.78, 3.03)	
Quartile 2	82	2.5 - 5.1	-0.71 (-1.96, 0.55)		-1.76 (-4.69, 1.17)	
Quartile 1	81	0.7 - 2.4	Referent		Referent	

Age 12-19

Quartile 4	171	11.2 - 549.2	-1.51 (-2.81, -0.21)	0.02	-2.18 (-4.99, 0.63)	0.1
Quartile 3	172	5.2 - 11.1	-2.14 (-3.54, -0.74)		-3.67 (-6.90, -0.44)	
Quartile 2	165	2.3 - 5.1	-1.38 (-2.59, -0.16)		-1.99 (-4.60, 0.64)	
Quartile 1	174	0.7 - 2.2	Referent		Referent	

Age 20-59

Quartile 4	192	8.4 - 392.5	-1.68 (-3.57, 0.21)	0.02	-2.17 (-5.99, 1.65)	0.08
Quartile 3	188	4.0 - 8.2	-1.32 (-2.86, 0.22)		-2.10 (-5.34, 1.13)	

Quartile 2	186	1.5 - 3.9	0.46 (-1.21, 2.14)		1.97 (-1.69, 5.64)	
Quartile 1	195	0.7 - 1.4	Referent		Referent	
Age 60-80						
Quartile 4	85	4.4 - 145.6	-2.07 (-3.42, -0.73)	0.01	-4.15 (-7.48, -0.81)	0.05
Quartile 3	87	2.3 - 4.3	-0.66 (-2.73, 1.42)		-0.73 (-5.52, 4.06)	
Quartile 2	62	1.0 - 2.2	-1.38 (-3.56, 0.80)		-1.89 (-5.85, 2.06)	
Quartile 1	114	0.7 - 0.8	Referent		Referent	
MEHHP	N	Range (mg/dL)	BMI β (95% CI)	p-trend	WC β (95% CI)	p-trend
Age 6-11						
Quartile 4	43	74.5 - 677.6	0.54 (-1.50, 2.57)	0.40	1.83 (-3.48, 7.13)	0.42
Quartile 3	43	36.8 - 74.4	1.60 (-0.53, 3.72)		3.14 (-2.41, 8.68)	
Quartile 2	43	17.1 - 36.5	0.64 (-1.30, 2.57)		1.63 (-2.96, 6.22)	
Quartile 1	43	0.7 - 16.4	Referent		Referent	
Age 12-19						
Quartile 4	85	55.0 - 2118.3	0.74 (-1.18, 2.65)	0.33	1.81 (-3.19, 6.83)	0.3
Quartile 3	87	28.7 - 54.7	1.45 (-0.11, 3.00)		3.84 (0.39, 7.29)	
Quartile 2	85	14.4 - 28.4	0.98 (-1.01, 2.97)		1.75 (-2.79, 6.29)	
Quartile 1	85	0.7 - 14.1	Referent		Referent	
Age 20-59						
Quartile 4	99	39.3 - 1521.4	1.08 (-0.75, 2.92)	0.29	3.13 (-0.73, 6.99)	0.09
Quartile 3	100	17.8 - 39.2	-0.97 (-3.17, 1.23)		-0.55 (-4.21, 3.10)	
Quartile 2	99	7.1 - 17.6	-0.28 (-2.14, 1.59)		-0.46 (-4.03, 3.10)	
Quartile 1	100	0.7 - 7.0	Referent		Referent	
Age 60-80						
Quartile 4	43	26.3 - 2227.5	-0.96 (-4.04, 2.11)	0.53	-2.82 (-8.89, 3.25)	0.38
Quartile 3	41	13.6 - 25.6	-1.93 (-5.11, 1.24)		-4.82 (-11.51, 1.86)	
Quartile 2	44	6.7 - 13.4	-1.24 (-3.53, 1.04)		-2.88 (-7.62, 1.86)	
Quartile 1	42	0.7 - 6.6	Referent		Referent	

MEOHP	N	Range (mg/dL)	BMI β (95% CI)	p-trend	WC β (95% CI)	p-trend
Age 6-11						
Quartile 4	43	50.5 - 687.0	-0.17 (-2.60, 2.26)	0.79	0.45 (-5.56, 6.46)	0.97
Quartile 3	43	26.4 - 49.4	0.52 (-1.44, 2.48)		0.72 (-4.99, 6.42)	
Quartile 2	43	11.6 - 24.7	0.61 (-1.30, 2.53)		1.52 (-3.28, 6.32)	
Quartile 1	43	0.8 - 11.1	Referent		Referent	
Age 12-19						
Quartile 4	86	39.1 - 1380.1	0.89 (-1.40, 3.18)	0.32	1.79 (-4.10, 7.68)	0.37
Quartile 3	85	20.4 - 38.6	0.99 (-0.79, 2.76)		2.62 (-1.64, 6.89)	
Quartile 2	86	10.7 - 20.3	0.43 (-1.74, 2.61)		0.49 (-4.40, 5.39)	
Quartile 1	85	0.8 - 10.5	Referent		Referent	
Age 20-59						

Quartile 4	99	28.0 - 914.2	0.38 (-1.90, 2.66)	0.62	1.52 (-2.98, 6.02)	0.38
Quartile 3	101	12.5 - 27.7	-0.77 (-3.08, 1.54)		-0.18 (-3.84, 3.48)	
Quartile 2	99	4.9 - 12.3	-1.03 (-2.28, 0.23)		-1.34 (-4.16, 1.49)	
Quartile 1	99	0.8 - 4.8	Referent		Referent	
Age 60-80						
Quartile 4	42	17.9 - 1026.4	0.94 (-2.98, 4.85)	0.8	2.46 (-7.41, 12.32)	0.71
Quartile 3	43	9.3 - 17.5	-0.76 (-3.70, 2.18)		-1.48 (-7.91, 4.95)	
Quartile 2	44	3.9 - 8.8	0.46 (-2.05, 2.97)		0.43 (-6.21, 7.07)	
Quartile 1	41	0.8 - 3.8	Referent		Referent	

* Adjusted for age, creatinine, height, race/ethnicity, socioeconomic status, % of daily calories from total fat (tertiles), daily servings of dairy (tertiles), daily servings of fruit and vegetables (tertiles), METS/month (continuous) (age 12+), TV/video/computer use (≤ 1 hour/day, >1 and <2.5 hours/day, ≥ 2.5 hours/day), smoking status (age 20+), and menopausal status and parity (women age 20+).

Appendix Table 2. Quartile ranges for 6 phthalate metabolites, and adjusted* mean difference in BMI (kg/m²) and WC (cm) by quartile, among males by age group, NHANES 1999-2002

MEP	N	Range (mg/dL)	BMI β (95% CI)	p-trend	WC β (95% CI)	p-trend
Age 6-11						
Quartile 4	82	194.4 - 9043.6	-0.02 (-1.49, 1.46)	0.65	-0.67 (-4.42, 3.09)	0.99
Quartile 3	83	75.6 - 191.7	0.97 (-0.27, 2.20)		1.42 (-1.73, 4.57)	
Quartile 2	82	39.3 - 75.5	-0.29 (-1.50, 0.92)		-0.75 (-4.01, 2.51)	
Quartile 1	82	4.7 - 39.0	Referent		Referent	
Age 12-19						
Quartile 4	165	525.9 - 12359.0	-0.13 (-1.63, 1.37)	0.89	-1.20 (-5.14, 2.74)	0.64
Quartile 3	166	195.6 - 524.9	0.02 (-1.13, 1.17)		-0.30 (-3.49, 2.89)	
Quartile 2	166	72.9 - 195.3	-0.05 (-1.23, 1.13)		-1.00 (-3.96, 1.96)	
Quartile 1	165	0.6 - 72.7	Referent		Referent	
Age 20-59						
Quartile 4	224	698.4 - 39938	0.82 (-0.15, 1.79)	0.11	2.19 (-0.51, 4.90)	0.11
Quartile 3	224	247.4 - 697.5	0.47 (-0.68, 1.63)		1.25 (-1.75, 4.25)	
Quartile 2	224	80.7 - 245.2	0.36 (-0.94, 1.67)		0.85 (-2.44, 4.15)	
Quartile 1	223	0.6 - 80.4	Referent		Referent	
Age 60-80						
Quartile 4	91	799.5 - 16995.2	1.05 (-0.10, 2.21)	0.03	1.68 (-1.75, 5.10)	0.21
Quartile 3	92	177.7 - 787.5	0.76 (-0.54, 2.06)		1.55 (-2.11, 5.21)	
Quartile 2	91	52.1 - 175.4	0.18 (-1.21, 1.56)		0.11 (-3.62, 3.85)	
Quartile 1	91	0.6 - 51.5	Referent		Referent	
MBP	N	Range (mg/dL)	BMI β (95% CI)	p-trend	WC β (95% CI)	p-trend
Age 6-11						
Quartile 4	82	71.4 - 638.5	0.80 (-0.42, 2.03)	0.56	1.25 (-1.91, 4.40)	0.86
Quartile 3	83	37.9 - 70.7	-0.24 (-1.91, 1.42)		-1.28 (-5.74, 3.18)	
Quartile 2	83	18.5 - 37.8	0.77 (-0.37, 1.90)		1.24 (-1.72, 4.19)	
Quartile 1	81	1.5 - 18.4	Referent		Referent	
Age 12-19						
Quartile 4	165	54.6 - 969.9	-0.87 (-2.54, 0.79)	0.2	-1.47 (-5.41, 2.48)	0.31
Quartile 3	166	31.2 - 54.4	-0.53 (-1.77, 0.70)		-0.70 (-4.02, 2.62)	
Quartile 2	166	16.5 - 31.1	0.09 (-1.32, 1.49)		0.83 (-2.78, 4.43)	
Quartile 1	165	0.6 - 16.4	Referent		Referent	
Age 20-59						
Quartile 4	223	41.6 - 397.1	0.65 (-0.39, 1.69)	0.11	2.91 (0.22, 5.60)	0.01
Quartile 3	226	23.4 - 41.3	1.22 (0.35, 2.09)		3.67 (1.27, 6.07)	
Quartile 2	224	11.3 - 23.3	0.66 (-0.48, 1.79)		1.86 (-1.05, 4.77)	
Quartile 1	222	0.6 - 11.3	Referent		Referent	

Age 60-80

Quartile 4	91	38.4 - 4363.4	-1.12 (-2.49, 0.24)	0.04	-2.60 (-6.05, 0.85)	0.08
Quartile 3	91	20.2 - 38.3	-1.44 (-2.61, -0.28)		-2.60 (-5.27, 0.07)	
Quartile 2	92	10.3 - 19.8	-0.36 (-1.79, 1.07)		-0.65 (-4.09, 2.80)	
Quartile 1	91	0.6 - 10.0	Referent		Referent	

MBzP	N	Range (mg/dL)	BMI β (95% CI)	p-trend	WC β (95% CI)	p-trend
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Age 6-11

Quartile 4	82	77.0 - 721.9	-0.13 (-1.53, 1.28)	0.80	0.55 (-3.31, 4.40)	0.85
Quartile 3	83	35.0 - 75.6	1.09 (-0.36, 2.54)		2.42 (-1.43, 6.27)	
Quartile 2	82	15.3 - 34.9	1.05 (-0.60, 2.71)		2.52 (-1.71, 6.74)	
Quartile 1	82	0.4 - 15.0	Referent		Referent	

Age 12-19

Quartile 4	166	49.7 - 1274.3	0.84 (-0.47, 2.15)	0.3	3.10 (-0.67, 6.88)	0.15
Quartile 3	164	26.0 - 49.5	0.21 (-0.85, 1.27)		1.36 (-1.47, 4.19)	
Quartile 2	166	12.0 - 25.9	0.60 (-0.65, 1.86)		2.14 (-0.99, 5.28)	
Quartile 1	166	0.2 - 11.8	Referent		Referent	

Age 20-59

Quartile 4	223	30.9 - 1197.6	2.35 (1.04, 3.65)	0.0002	6.63 (3.42, 9.84)	<.0001
Quartile 3	223	14.8 - 30.8	1.70 (0.65, 2.76)		4.87 (2.18, 7.56)	
Quartile 2	226	7.4 - 14.6	0.47 (-0.53, 1.48)		1.27 (-1.34, 3.87)	
Quartile 1	223	0.2 - 7.3	Referent		Referent	

Age 60-80

Quartile 4	91	25.3 - 414.8	-1.59 (-3.43, 0.24)	0.06	-3.18 (-7.64, 1.28)	0.09
Quartile 3	93	11.3 - 25.0	-1.27 (-2.97, 0.42)		-1.84 (-5.61, 1.93)	
Quartile 2	90	4.8 - 11.2	-0.35 (-1.60, 0.89)		-0.11 (-3.65, 3.43)	
Quartile 1	91	0.2 - 4.7	Referent		Referent	

MEHP	N	Range (mg/dL)	BMI β (95% CI)	p-trend	WC β (95% CI)	p-trend
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Age 6-11

Quartile 4	82	11.7 - 196.3	-0.22 (-1.32, 0.89)	0.76	-0.20 (-2.98, 2.57)	0.96
Quartile 3	82	5.2 - 11.4	0.25 (-0.84, 1.35)		0.39 (-2.76, 3.53)	
Quartile 2	84	2.2 - 5.1	0.14 (-1.06, 1.33)		0.06 (-2.99, 3.11)	
Quartile 1	81	0.7 - 2.1	Referent		Referent	

Age 12-19

Quartile 4	167	8.6 - 273.4	-0.50 (-1.95, 0.94)	0.46	-1.39 (-5.15, 2.37)	0.44
Quartile 3	166	3.7 - 8.5	-1.32 (-2.58, -0.06)		-3.43 (-6.86, -0.01)	
Quartile 2	165	1.8 - 3.6	-0.97 (-2.24, 0.31)		-2.45 (-5.82, 0.91)	
Quartile 1	164	0.7 - 1.7	Referent		Referent	

Age 20-59

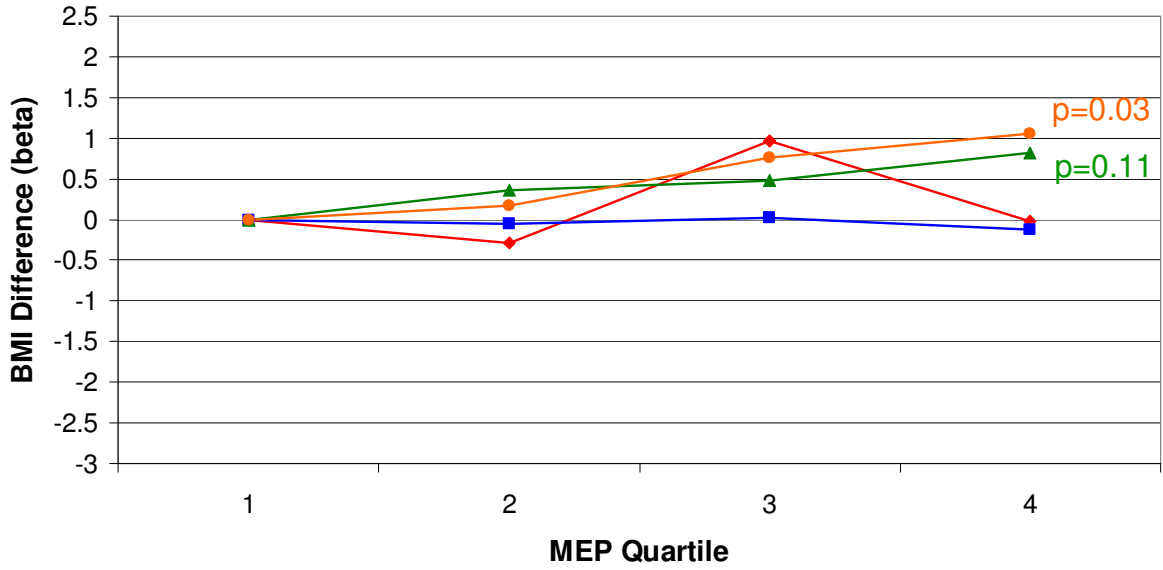
Quartile 4	223	10.0 - 615.6	0.44 (-0.63, 1.52)	0.35	0.91 (-1.43, 3.24)	0.44
Quartile 3	220	4.6 - 9.8	0.26 (-1.05, 1.57)		0.02 (-3.39, 3.43)	

Quartile 2	231	1.9 - 4.5	-0.04 (-0.92, 0.83)		-0.15 (-2.47, 2.17)	
Quartile 1	221	0.7 - 1.8	Referent		Referent	
Age 60-80						
Quartile 4	90	5.9 - 204.2	-1.16 (-2.60, 0.28)	0.08	-2.42 (-5.76, 0.93)	0.16
Quartile 3	91	2.5 - 5.6	-0.69 (-2.16, 0.79)		-0.57 (-4.27, 3.12)	
Quartile 2	78	1.0 - 2.4	0.11 (-1.39, 1.61)		0.64 (-3.16, 4.45)	
Quartile 1	106	0.7 - 0.8	Referent		Referent	
MEHHP	N	Range (mg/dL)	BMI β (95% CI)	p-trend	WC β (95% CI)	p-trend
Age 6-11						
Quartile 4	48	66.4 - 622.7	0.42 (-1.09, 1.92)	0.57	1.27 (-2.43, 4.96)	0.40
Quartile 3	49	34.4 - 65.2	0.27 (-0.97, 1.52)		1.11 (-2.44, 4.65)	
Quartile 2	49	19.1 - 34.3	0.30 (-1.01, 1.61)		0.45 (-3.37, 4.26)	
Quartile 1	48	0.7 - 18.7	Referent		Referent	
Age 12-19						
Quartile 4	80	54.9 - 977.0	1.00 (-0.69, 2.69)	0.13	2.15 (-1.77, 6.08)	0.2
Quartile 3	81	27.1 - 54.2	2.37 (0.62, 4.11)		6.53 (2.23, 10.82)	
Quartile 2	80	12.9 - 26.9	0.94 (-0.57, 2.46)		3.07 (-0.06, 6.19)	
Quartile 1	81	0.7 - 12.5	Referent		Referent	
Age 20-59						
Quartile 4	125	48.8 - 3236.1	1.74 (-0.28, 3.76)	0.1	4.60 (-0.03, 9.24)	0.08
Quartile 3	126	21.6 - 48.7	0.35 (-1.17, 1.87)		0.65 (-3.23, 4.52)	
Quartile 2	126	11.0 - 21.4	0.06 (-1.33, 1.46)		0.80 (-2.67, 4.28)	
Quartile 1	125	0.7 - 10.9	Referent		Referent	
Age 60-80						
Quartile 4	46	30.8 - 1408.9	0.41 (-2.47, 3.28)	0.85	0.68 (-7.42, 8.78)	0.83
Quartile 3	47	12.9 - 30.5	-0.84 (-3.84, 2.16)		-0.95 (-8.84, 6.94)	
Quartile 2	46	7.1 - 12.8	-0.25 (-2.59, 2.09)		-1.10 (-6.97, 4.78)	
Quartile 1	47	0.7 - 6.3	Referent		Referent	
MEOHP	N	Range (mg/dL)	BMI β (95% CI)	p-trend	WC β (95% CI)	p-trend
Age 6-11						
Quartile 4	48	47.2 - 351.6	0.14 (-1.21, 1.48)	0.91	0.60 (-2.68, 3.88)	0.77
Quartile 3	50	22.9 - 46.5	-0.09 (-0.99, 0.80)		0.06 (-2.55, 2.66)	
Quartile 2	47	13.0 - 22.6	0.17 (-1.36, 1.70)		0.74 (-3.95, 5.42)	
Quartile 1	49	0.8 - 12.8	Referent		Referent	
Age 12-19						
Quartile 4	81	36.1 - 607.1	0.27 (-1.40, 1.94)	0.65	0.68 (-2.67, 4.02)	0.76
Quartile 3	79	18.5 - 35.8	1.74 (-0.08, 3.56)		4.67 (0.11, 9.23)	
Quartile 2	82	8.8 - 18.3	0.92 (-1.14, 2.97)		3.34 (-1.09, 7.78)	
Quartile 1	80	0.8 - 8.7	Referent		Referent	
Age 20-59						

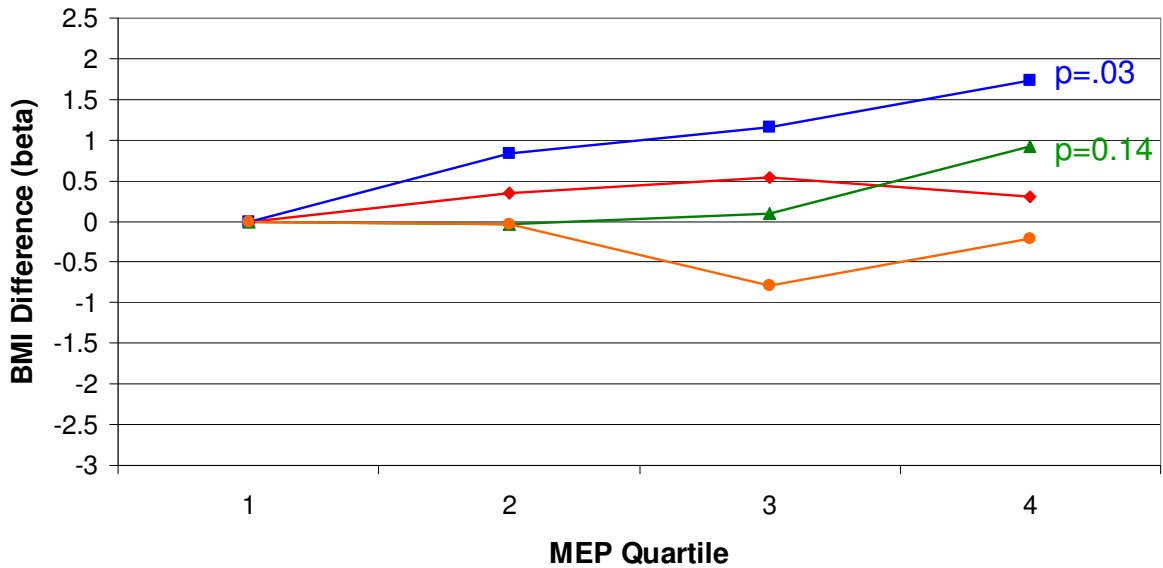
Quartile 4	125	31.5 - 926.7	2.14 (-0.13, 4.41)	0.09	5.81 (0.69, 10.94)	0.06
Quartile 3	126	14.7 - 30.7	0.96 (-0.67, 2.60)		1.78 (-1.98, 5.54)	
Quartile 2	125	7.0 - 14.6	0.65 (-0.77, 2.07)		2.18 (-1.15, 5.51)	
Quartile 1	126	0.8 - 6.9	Referent		Referent	
Age 60-80						
Quartile 4	46	19.9 - 766.9	0.69 (-2.05, 3.44)	0.75	2.31 (-4.97, 9.59)	0.6
Quartile 3	48	9.1 - 19.7	-0.94 (-3.42, 1.55)		-1.02 (-7.79, 5.76)	
Quartile 2	46	4.9 - 9.0	0.03 (-1.94, 2.01)		0.22 (-4.93, 5.38)	
Quartile 1	46	0.8 - 4.8	Referent		Referent	

* Adjusted for age, creatinine, height, race/ethnicity, socioeconomic status, % of daily calories from total fat (tertiles), daily servings of dairy (tertiles), daily servings of fruit and vegetables (tertiles), METS/month (continuous), TV/video/computer use (\leq 1 hour/day, >1 and <2.5 hours/day, \geq 2.5 hours/day), and smoking status (age 20+)

1A: Males



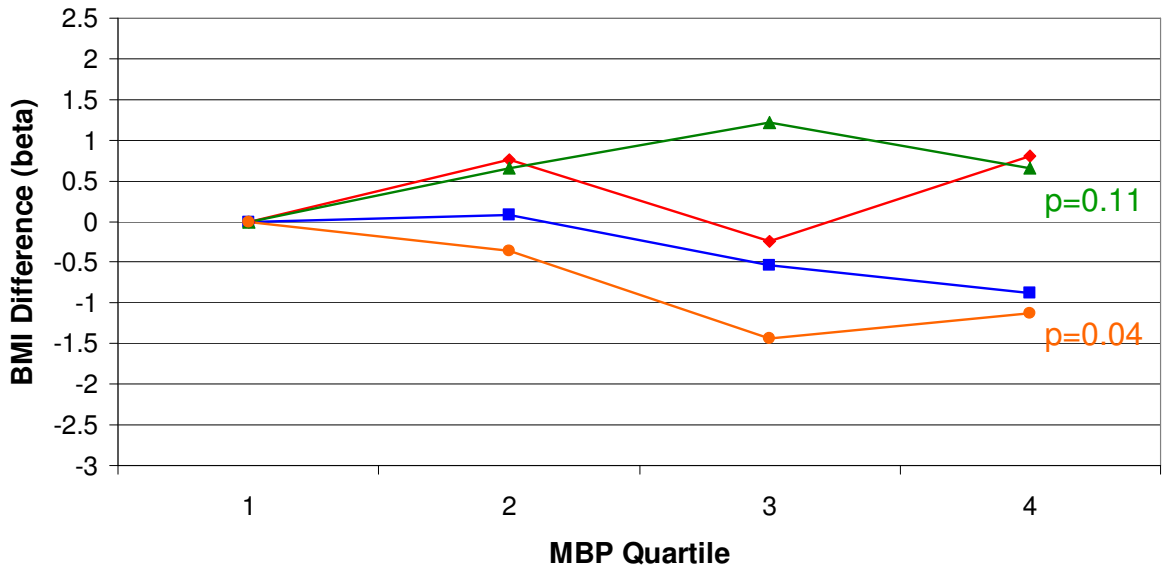
1B: Females



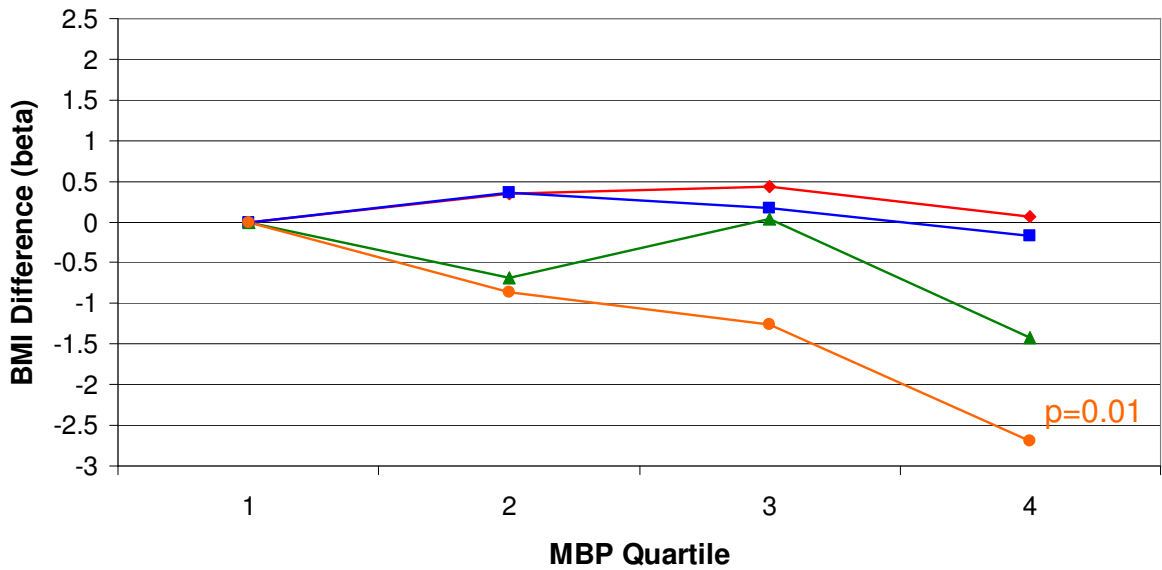
◆ 6 to 11 years ■ 12 to 19 years ▲ 20 to 59 years ● 60 to 80 years

Figure 1

2A: Males

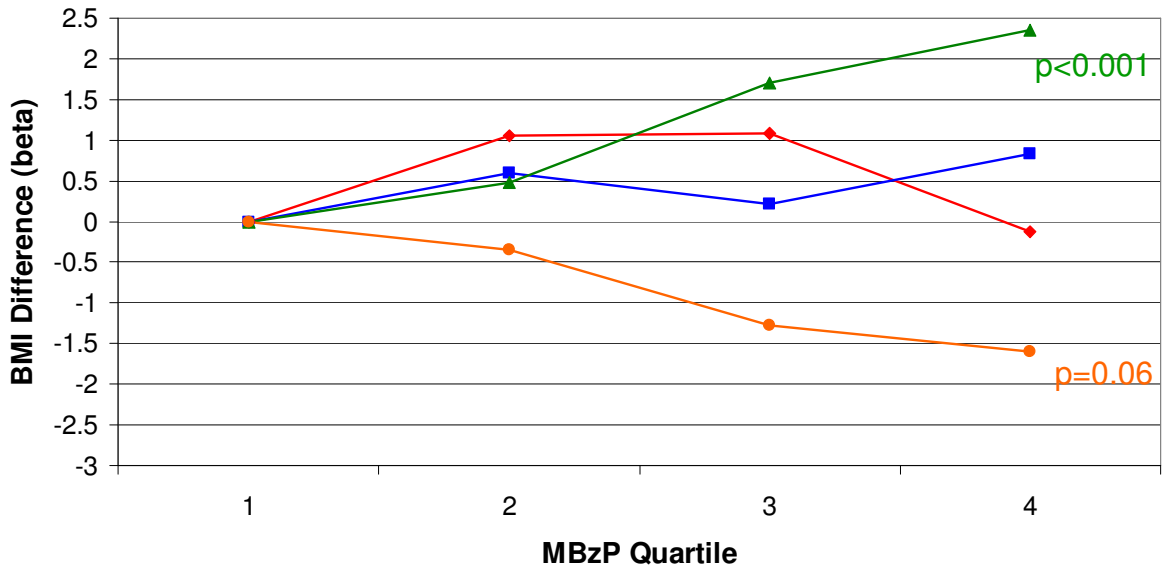


2B: Females

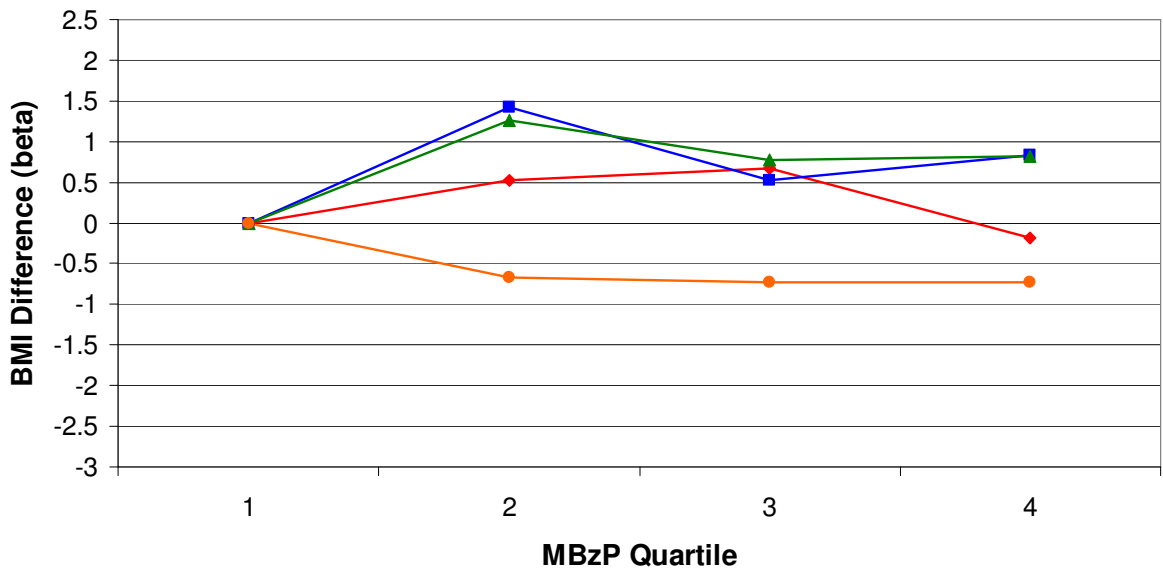


◆ 6 to 11 years ■ 12 to 19 years ▲ 20 to 59 years ● 60 to 80 years

3A: Males



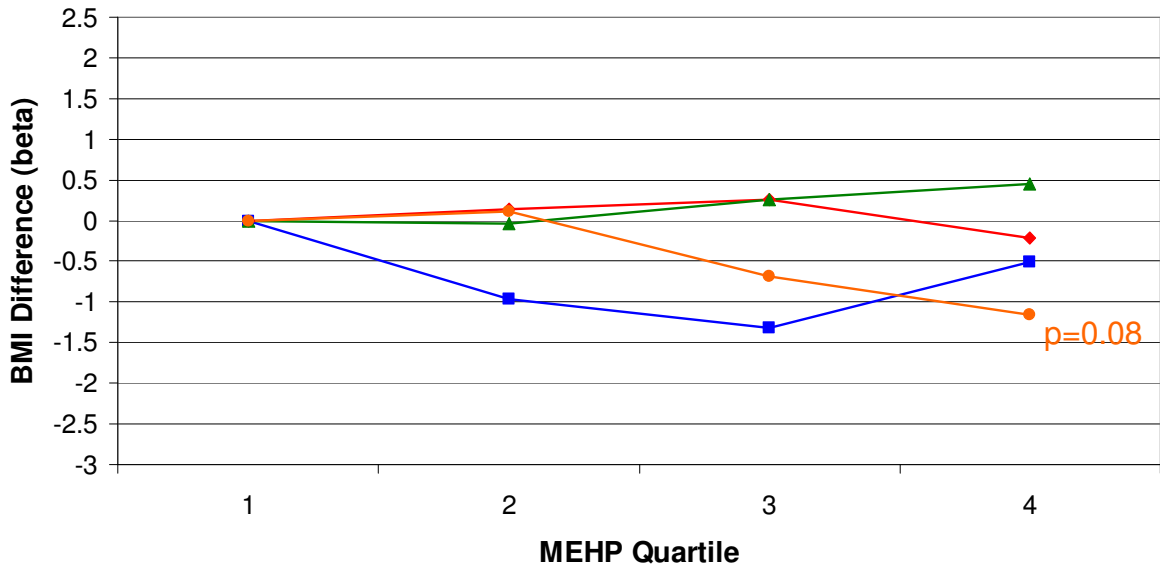
3B: Females



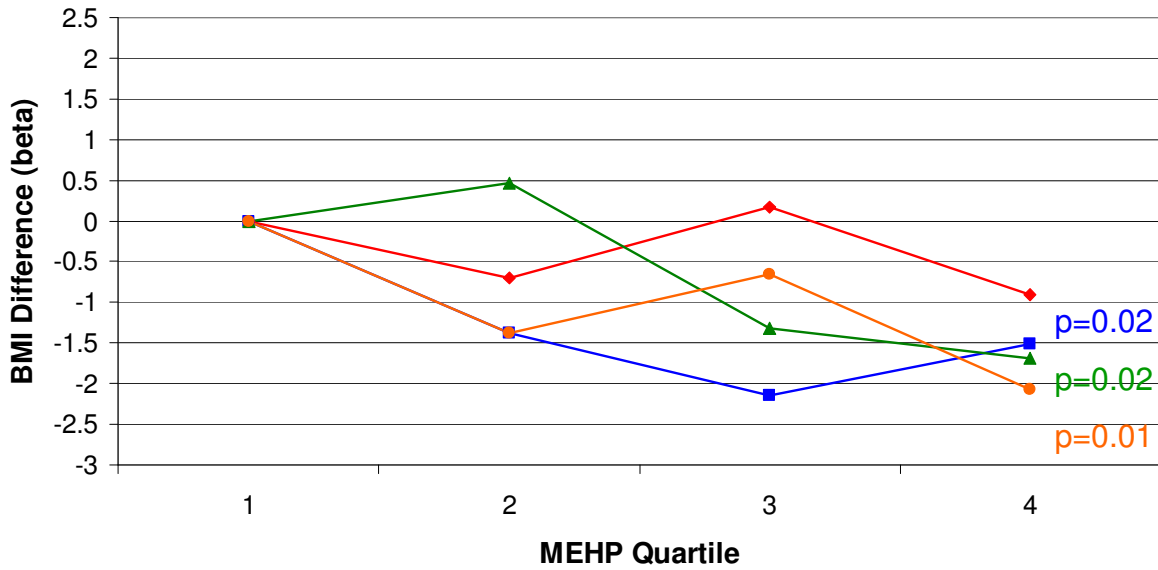
◆ 6 to 11 years ■ 12 to 19 years ▲ 20 to 59 years ● 60 to 80 years

Figure 3

4A: Males



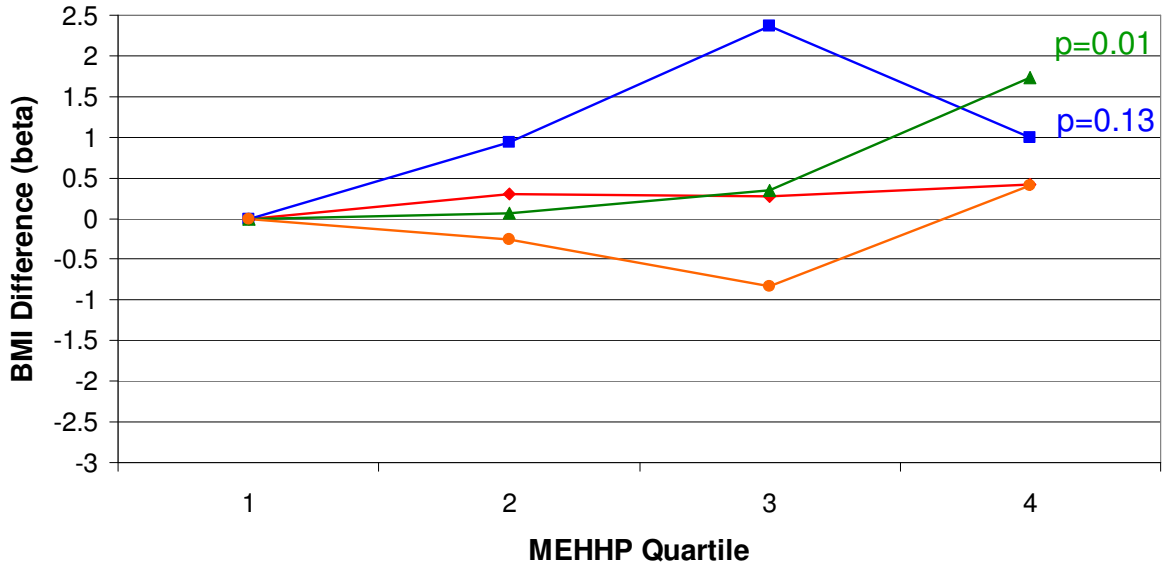
4B: Females



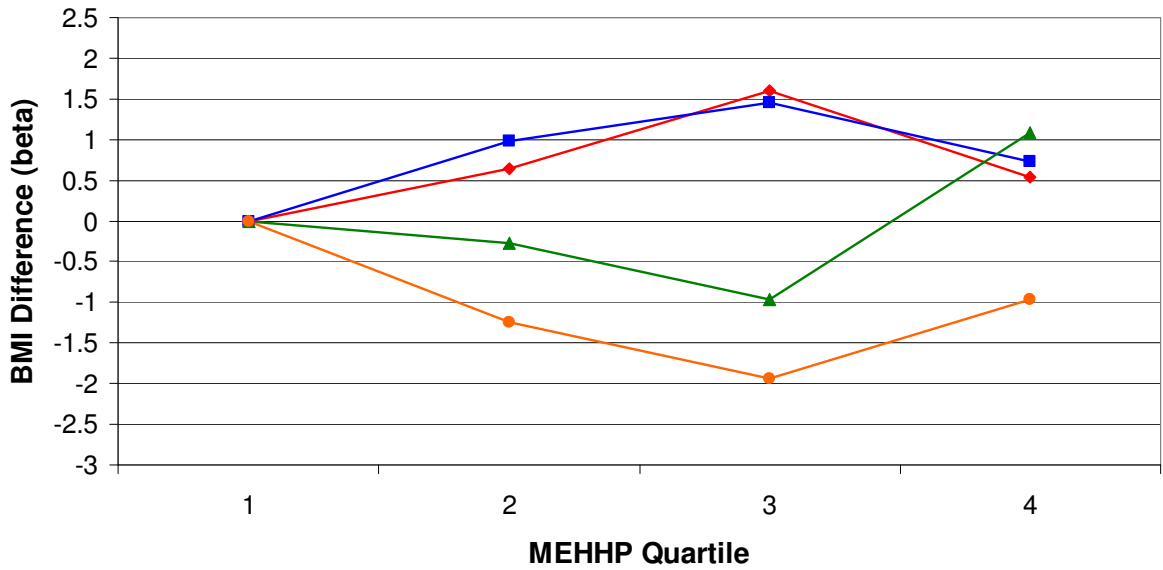
◆ 6 to 11 years ■ 12 to 19 years ▲ 20 to 59 years ● 60 to 80 years

Figure 4

5A: Males



5B: Females



◆ 6 to 11 years ■ 12 to 19 years ▲ 20 to 59 years ● 60 to 80 years

Figure 5