

ORIGINAL ARTICLE

Assessment of chemical coexposure patterns based upon phthalate biomonitoring data within the 2007/2008 National Health and Nutrition Examination Survey

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As regulatory initiatives increasingly call for an understanding of the cumulative risks from chemical mixtures, evaluating exposure data from large biomonitoring programs, which may inform these cumulative risk assessments, will improve the understanding of occurrence and patterns of coexposures. Here we have analyzed the urinary metabolite data for six phthalates (di-butyl phthalate; di-isobutyl phthalate; butyl-benzyl phthalate; bis(2-ethylhexyl) phthalate; di-isononyl phthalate; and di-isodecyl phthalate) in the 2007/2008 National Health and Nutrition Examination Survey (NHANES) data set. For the total data set ($N = 2604$), the co-occurrence of multiple phthalates at the upper percentile of exposure was infrequent. There were no individuals in the NHANES sample who were exposed to >95th percentiles for all six phthalates. For 75% of individuals, none of the six phthalates were above the 95th percentile of their respective exposure distributions. These data suggest that high exposure to multiple phthalates is infrequent in the NHANES population. This analysis solely focused on the pattern of contribution of individual phthalates to total exposure. It did not address the pattern of contribution to potential risk. The approach presented could potentially be used to provide insight into understanding the coexposure patterns for other chemicals.

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INTRODUCTION

As an integral component of the risk assessment process, exposure assessment is typically conducted from the perspective of the single chemical using conservative exposure levels (i.e. it is intentionally designed to represent relatively higher exposure). For example, typically the “reasonable maximum exposure” (RME) is used as “the highest exposure that could reasonably be expected to occur for a given exposure pathway at a site”.^{1,2} In fact, the Environmental Protection Agency (EPA)³ advised that the RME from the upper end of the range of risk estimates should be used for probabilistic risk assessment, which typically forms the basis of EPA decision-making. In practice, the 95th percentile value is commonly used for the RME for a single chemical assessment. Assessment science is still emerging for coexposure.^{4,5} An improved understanding on how the upper percentile value for a single chemical exposure distribution contributes to the coexposure patterns when multiple chemicals are examined can be very useful.

Increased interest has been placed on cumulative risk assessment or the estimate of risk from exposure to multiple chemicals (chemical coexposures).^{4,6–9} Cumulative risk assessment presents a challenge, because limited information is available on patterns of chemical coexposures. For example, if considering two substances, coexposure may be unrelated (independent), positively correlated (higher exposure to one predicts higher exposure to the other), or negatively correlated (higher exposure to one predicts lower exposure to the other). Exposure modeling tools, typically designed for single chemical assessments, require

additional information to understand how coexposure estimates should be developed.

The understanding of chemical exposures has been greatly enhanced by the availability of general population-based human biomonitoring data since these data allow for the determination of the internal dose and external exposures from resulting measurements of chemicals, their metabolites, or reaction products in biologic media (e.g. urine or blood). Biomonitoring data represent an integrated measure of exposure from multiple sources and routes, and can be used to assess total exposure to the measured substances. When biomonitoring studies are designed such that multiple biomarkers are quantified in individual samples, the results can also be used to provide insights to understanding patterns of coexposures to multiple chemicals. One such category of chemicals that has been examined in biomonitoring programs is the phthalate esters chemical family.

Phthalates, which are dialkyl or alkyl aryl esters of phthalic acid, are used in a wide variety of applications including cosmetics, consumer products including personal-care products, pharmaceuticals, medical devices, children’s toys, food packaging, cleaning, and building materials.¹⁰ Because of their physical/chemical properties, the end-use patterns for various phthalates can be quite different. For example, the high-molecular-weight phthalates (di-isononyl phthalate, (DINP) and di-isodecyl phthalate (DIDP)) are commonly used as plasticizers in PVC polymers and plastisol applications (e.g. building materials like flooring, wire and cabling, car interior), whereas the low-molecular-weight phthalates (di-butyl phthalate (DBP), di-isobutyl phthalate (DiBP),

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butyl-benzyl phthalate (BBP), and bis(2-ethylhexyl) phthalate (DEHP) are used in non-PVC applications (e.g. personal-care products, paints)¹¹ (Supplementary Table 1).

Regardless of the differences in end-use patterns, it is recognized that there is ubiquitous exposure to phthalates because of their identification in multiple human biomonitoring programs. However, what has not yet been investigated from the data these programs have generated is an understanding of the coexposure patterns present within the sample populations and how this information could be interpreted when assessing the overall exposure burden for these classes of substance. The National Health and Nutrition Examination Survey (NHANES) data set provides results of multiple phthalate metabolites by individual, and thus can provide insight into patterns of phthalate coexposure. Here, we have analyzed the urinary metabolite data for six phthalates commonly used in consumer products (DBP, DiBP, BBP, DEHP, DINP, and DIPP) from the 2007/2008 NHANES data set to better understand phthalate coexposure patterns within various demographics represented in the data.

METHODS

Data Source

Urinary phthalate metabolite data were obtained from the NHANES 2007/2008 data set. NHANES is a cross-sectional survey designed to monitor the health and nutritional status of the civilian non-institutionalized US population (<http://www.cdc.gov/nchs/nhanes.htm>). The survey collects data through interviews, standardized physical examinations, and laboratory tests using blood and urine specimens provided by participants.

Six phthalates were chosen in this analysis from the NHANES 2007/2008 data set owing to their use in wide range of consumer products in the United States (Supplementary Table 1). The data for 10 urinary phthalate metabolites of the six phthalates that represent the majority of the market share were retrieved: mono-*n*-butyl phthalate for DBP, mono-isobutyl phthalate for DiBP, monobenzyl phthalate for BBP, mono(2-ethylhexyl) phthalate, mono(2-ethyl-5-hydroxyhexyl) phthalate, mono(2-ethyl-5-oxohexyl) phthalate, and mono(2-ethyl-5-carboxypentyl) phthalate for DEHP, mono-isononyl phthalate and mono-carboxyisooctyl phthalate for DINP, and mono-carboxyisooctyl phthalate for DIDP.

Estimating Daily Intake

Intake estimates of phthalate esters were calculated from their creatinine corrected spot urinary metabolite levels using Eq. (1).^{12, 13} As described in the NHANES protocol, in instances where sample concentrations were below the limit of detection (LOD) (i.e. non-detects), the value was replaced by the LOD divided by the square root of 2:

$$DI = \frac{UC \times CE}{F_{UE} \times 1000} \times \frac{MW_d}{MW_m} \tag{1}$$

DI is daily intake ($\mu\text{g}/\text{kg}/\text{day}$), UC is the creatinine corrected urinary metabolite concentration ($\mu\text{g}/\text{g}$), CE is the creatinine excretion rate ($\text{mg}/\text{kg}/\text{day}$) for adults¹⁴ and children¹⁵ and is used to account for differences in urine dilution,¹⁶ and F_{UE} is the fractional urinary excretion rate of the metabolite (unitless). MW_d and MW_m are the molecular weights of parent phthalate and the metabolite, respectively (values can be found in Table 1). In instances where multiple metabolites are examined (e.g. DEHP), the estimated daily intake is calculated as

$$DI = \sum_{i=1}^j DI_i \times \frac{F_{UEi}}{\sum_{i=1}^j F_{UEi}} \tag{2}$$

where i is the i th metabolite of a phthalate, j is the number of metabolites for a phthalate, and F_{UEi} is the fractional urinary excretion rate of the i th phthalate metabolite (unitless).

Percentile/Percentile Rank

Exposure distributions were also developed for the daily intake dose of each individual phthalate in the NHANES 2007/2008 data set. Each daily intake value in the exposure distribution can be characterized by a percentile rank. The percentile rank is the percentage (e.g. 95) of data points that falls at or below a certain value. The percentile, in contrast, is the smallest value of a variable (e.g. concentration), with the property that

Table 1. Molecular weights, detection limits, and fractional urinary excretion rate (F_{UE}) of the metabolites of six phthalate diesters examined in the 2007/2008 NHANES data set.

Phthalate diester metabolite	Molecular weight	LOD ^a ($\mu\text{g}/\text{l}$)	Number of individuals < LOD	F_{UE}
DBP	278.34			
MBP	222.24	0.6	19	0.69 ^b
DiBP	278.34			
MiBP	222.24	0.3	47	0.69 ^b
BBP	312.35			
MBzP	256.25	0.216	46	0.73 ^b
DEHP	390.57			
MEHP	278.35	1.1	858	0.062 ^c
5OH-MEHP (MEHHP)	294.34	0.7	20	0.149 ^c
5oxo-MEHP (MEOHP)	292.33	0.6	44	0.109 ^c
5cx-MEPP (MECPP)	308.33	0.5	2	0.132 ^c
DINP	420.6			
MINP	292.37	1.232	2320	0.03 ^d
7cx-MMeHP (MCOP)	322.35	0.7	94	0.099 ^d
DIDP	446.68			
7cx-MMeOP (MCNP)	336.38	0.5	249	0.099 ^{d,e}

Abbreviations: BBP, butyl-benzyl phthalate; DBP, di-butyl phthalate; DEHP, bis(2-ethylhexyl) phthalate; DiBP, di-isobutyl phthalate; DIDP, di-isodecyl phthalate; DINP, di-isononyl phthalate; F_{UE} , fractional urinary excretion rates; LOD, limit of detection; MBP, mono-*n*-butyl phthalate; MBzP, monobenzyl phthalate; MCNP, mono-carboxyisooctyl phthalate; MCOP, mono-carboxyisooctyl phthalate; MECPP, mono(2-ethyl-5-carboxypentyl) phthalate; MEHP, mono(2-ethylhexyl) phthalate; MEHHP, mono(2-ethyl-5-hydroxyhexyl) phthalate; MEOHP, mono(2-ethyl-5-oxohexyl) phthalate; MiBP, mono-isobutyl phthalate; MINP, mono-isononyl phthalate; NHANES, National Health and Nutrition Examination Survey.

^aInstances where the sample concentrations were below LOD (i.e. non-detects) were replaced by the LOD divided by the square root of 2.

^b F_{UE} reported in Anderson et al.²⁸

^c F_{UE} reported in Koch et al.²⁹

^d F_{UE} reported in Anderson et al.³⁰ and Koch and Angerer.³¹

^eValue used was reported data for DINP as described by Kransler et al.²¹

$n\%$ of the data values are less than or equal to it. The daily intake doses and relevant percentile ranks for each of the six phthalates calculated were included in the Supplementary Material (Supplementary Data set).

Study Population

We analyzed the coexposure patterns of the six phthalates for the sample population, and also the subsets of the sample population stratified by age (pre-reproductive: 6–11; reproductive: 12–40; and post-reproductive: 40+), gender (female and male), and ethnicity (White, Hispanic, Black) as these factors have been indicated to have potential influence on phthalate exposure levels.¹⁷

Statistical Analysis

The NHANES sample is selected through a complex, multistage process, including oversampling of ethnic minorities and young children, and provided with sampling weight. The sample weight takes into account unequal probabilities of selection resulting from the complex sample design, non-response, and non-coverage, and thus can generate results representative for the US general population. In this analysis, the weighting

was applied in developing the exposure distribution of the six phthalate esters on both the sample and subsample (age 6–11 years, female and male) using SAS software version 9.3 (SAS Institute, Cary, NC, USA) and SUDAAN software version 11.0 (RTI, Research Triangle Park, NC, USA). The percentile was calculated using the SAS PROC SURVEYMEANS procedure.

RESULTS

The complete 2007/2008 NHANES data set, which contains phthalate metabolite data for 2604 individuals, was used for this analysis. The sample consisted of 1294 males and 1310 females aged 6 years or older, of whom 41% ($n = 1077$) were non-Hispanic White, 32% ($n = 833$) were Hispanic, 23% ($n = 597$) were non-Hispanic black, and 4% ($n = 97$) were other race including multiple races (Table 2). The number of individuals with phthalate metabolite below its LOD was listed (Table 1).

Daily intake percentiles of the six phthalates for the US population from the NHANES 2007/2008 data set are provided (Table 3). The focus of this analysis, however, is on patterns of coexposure rather than on absolute values. The relative contribution of each phthalate to the summed total exposure of each sampled individual is presented for both the midpoints (Figure 1a) and the upper percentiles (Figure 1b) of the summed exposure. At the 50th percentile, it can be seen that multiple phthalates contribute to the overall exposure, whereas at the >99th percentile exposure (used as an example of upper percentile exposure) is typically dominated by a single phthalate.

The coexposure patterns of the six phthalates were further explored using the percentile values of each individual phthalate exposure distribution (Figure 2). For the total data set ($N = 2604$), the co-occurrence of multiple phthalates, each being at the upper percentile of exposure for that specific phthalate, was infrequent (Figure 2a). There were no individuals who were >95th percentile distribution for all six phthalates examined. In only three individuals (0.12%), five phthalates fell above the 95th percentiles. For 75% of individuals, none of the six phthalates were above the 95th percentile of their respective exposure distributions. The number of phthalates above a given percentile is more evenly distributed for lower exposure percentiles (Figures 2b–d). For example, 17% of individuals had all six phthalates above the 50th percentile, and 12% of individuals had none of the six phthalate above the 50th percentile (Figure 2d).

A scatter plot was also created to illustrate the extent to which the coexposure patterns of the six phthalates vary with the level of total phthalate exposure (Figure 3). For each individual sampled, the total phthalate exposure was calculated as the sum of the exposure for each of the six phthalates and the total percentile rank was calculated as the sum of the percentile rank of each phthalate. The rank of the total exposure was plotted against the rank of the total exposure percentiles for each individual, demonstrating that the scatter is greatest at the higher levels of total exposure (Figure 3). This indicates that the highest phthalate total exposures occur through a variety of coexposure patterns, and are not necessarily dominated by the contribution of the upper percentiles of the exposure distributions for each individual phthalate.

A cumulative distribution of the averaged exposure percentile ranks across the six phthalates was developed to explore what percentile rank of the individual phthalate exposure distributions, on average, would provide a summed coexposure estimate related to various percentile ranks of the population based upon the NHANES data set (Figure 4). Results indicate that 95% of individuals have total exposures that fall below an averaged percentile of 82%.

Similar coexposure patterns were also observed in the subsample populations: age 6–11 years (Figure 5), female-all age or male-all age from the weighted analysis described above (Supplementary Figure 1). In addition, we conducted unweighted

Table 2. Sample demographics within the 2007/2008 NHANES data set.

	Count	Mean
<i>Age (years)</i>		
6–11	389	9
12–40	1023	25
40 +	1192	61
Total	2604	39
	Count	Percentage
<i>Race</i>		
White	1077	41
Hispanic	833	32
Black	597	23
Other	97	4
<i>Gender</i>		
Female	1310	50
Male	1294	50

Abbreviation: NHANES, National Health and Nutrition Examination Survey.

Table 3. Selected percentiles of estimated daily intake ($\mu\text{g}/\text{kg}/\text{day}$) for six phthalates examined from the 2007/2008 NHANES data set

	5%	10%	25%	50%	75%	90%	95%
DBP	0.12	0.18	0.29	0.54	0.98	1.65	2.43
DiBP	0.04	0.06	0.12	0.22	0.38	0.60	0.92
BBP	0.04	0.06	0.13	0.26	0.58	1.14	1.69
DEHP	0.56	0.72	1.14	2.09	4.28	10.12	20.38
DINP	0.29	0.39	0.61	1.11	2.34	5.28	9.36
DIDP	0.11	0.16	0.26	0.47	0.92	1.75	2.84

Abbreviations: BBP, butyl-benzyl phthalate; DBP, di-butyl phthalate; DEHP, bis(2-ethylhexyl) phthalate; DiBP, di-isobutyl phthalate; DIDP, di-isodecyl phthalate; DINP, di-isononyl phthalate; NHANES, National Health and Nutrition Examination Survey.

Values were calculated using sample data and Eq. (1)

analysis (without applying sample weight) for subpopulations stratified by age (6–11, 12–40, and 41 + years), gender (female), and ethnicities (White, Hispanic, and Black), and also observed similar coexposure patterns (Supplementary Figure 2).

DISCUSSION

Assessment of coexposures is not currently well established as a risk assessment approach and presents unique challenges. Exposure models typically predict exposure to individual substances. Predicting coexposures requires integrated knowledge of multiple exposure sources for multiple substances and the likelihood of contact with these sources. For example, information important to consider includes: (1) use patterns of individual products and how product use may be correlated, that is to say if an individual used product A are they more or less likely to use product B, (2) functionality of product ingredients in formulated products (if two ingredients have the same function, are they likely to be used together or do they preclude each other's use), and (3) human behaviors that affect cause or exposure patterns (direct product use and/or indirect contact with substances in environmental media). Therefore, an analysis on coexposure pattern could be more informative than using the high end of exposure values for single substances as conservative estimates.

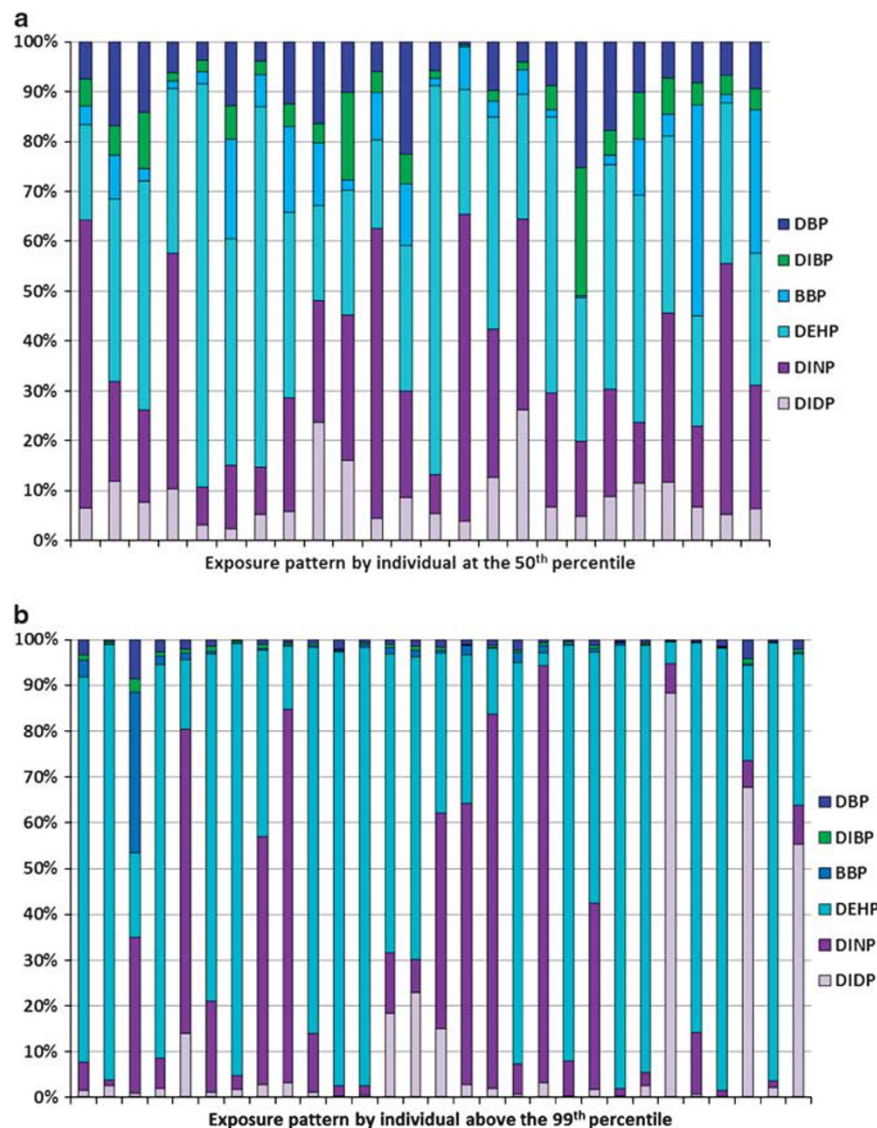


Figure 1. Percent contribution of individual phthalates to the sum-total exposure for individuals at the 50th and greater than the 99th percentile. The relative percent contributions of the six individual phthalates examined are presented for (a) individuals at the 50th percentile total exposure point and (b) individuals >99th percentile point on the sum-total estimated daily intake exposure distribution curve. At the 50th percentile, an individual's phthalate intake does not appear to be predominately the result of one phthalate. Conversely, individuals with the highest sum-total phthalate intake typically have one phthalate that contributes to a majority of the exposure.

Given the increasing complexity in assessing coexposures as compared with a single exposure, it is important to develop approaches that can help reduce uncertainty in the coexposure estimate. RMEs are typically used for conservative estimates of a single substance, but understanding of how to develop a reasonable maximum coexposure estimate is limited. The work here explores how biomonitoring data can be used to develop a realistic estimate of coexposures.

When biomonitoring data are available for multiple substances for a given individual, these data provide a direct approach for evaluating patterns of coexposure without the need to address the considerations previously mentioned. There exist a few large-scale surveys that include biomonitoring data for multiple phthalates, such as NHANES (<http://www.cdc.gov/nchs/nhanes.htm>), DEMOCOPHES in the European Union (<http://www.eu-hbm.info/democophes>), and Canadian health measures survey in Canada (<http://www.hc-sc.gc.ca/ewh-semt/contaminants/human-humaine/chms-ecms-eng.php>).

The NHANES data set, which includes data for multiple phthalates for given individuals, provides useful insights to coexposure patterns. The analysis on the six phthalates indicates that higher total exposures tended to be dominated by one phthalate. Coexposure to multiple phthalates at levels greater than the 95th percentile of individual phthalate exposure distributions did not frequently occur. Similar observations have been made for pesticides, which demonstrate that the probability of being jointly exposed to high values for all pesticides is very low,^{18,19} and co-occurrence is not random.²⁰

Similar analyses would be needed to see if the NHANES coexposure patterns are typical of other data sets. The NHANES data set is likely a good source of this information, as the data are designed to be representative of the US population and are consistent with other study data human biomonitoring data on phthalates.^{21–25} However, the NHANES survey does not provide information on the sources of phthalate exposure in the sample population or product use patterns, which limits its use

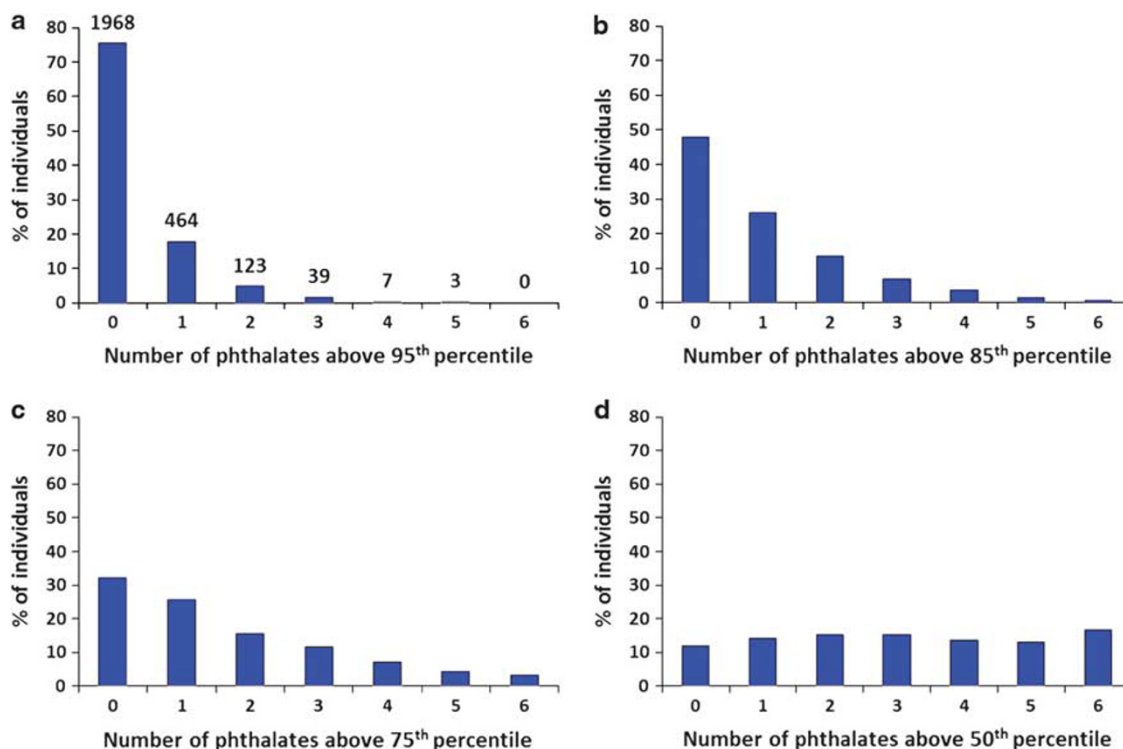


Figure 2. Percentage of individuals (all) with the number of phthalates above the 95th (a), 85th (b), 75th (c), and 50th (d) percentile exposure sample distribution. Exposure distributions were calculated for each of the six phthalates examined for the study sample. The data were then tabulated to show the percentage of individuals within the study sample that had an estimated phthalate exposure greater than the given percentile of the exposure sample distribution.

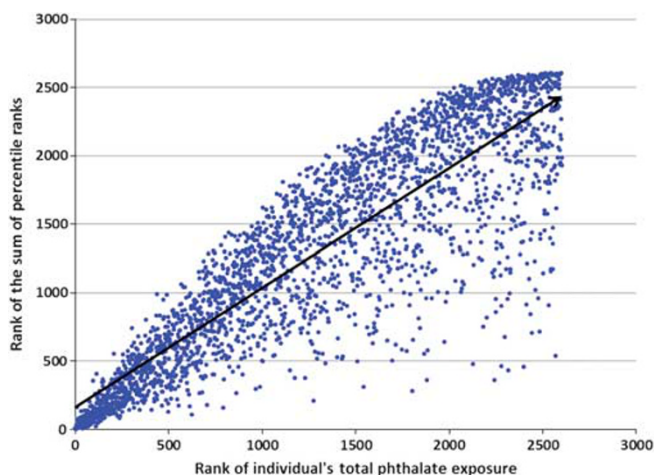


Figure 3. Scatter plot illustrating the greatest coexposure patterns at the highest total phthalate exposure. For each individual, the total exposure was calculated as the sum of exposure for each of the six phthalates and the total percentile rank was calculated as the sum of the percentile rank of each phthalate. The rank of the total exposure was plotted against the rank of the total percentile rank for each individual, demonstrating that the scatter is greatest at the upper percentile.

in linking the phthalate human exposure to their sources. In the future, collecting additional contextual information in conjunction with biomonitoring samples, such as behavioral information (product use, dietary intake, activity patterns) and maintaining this in a way that enables a more direct linkage with

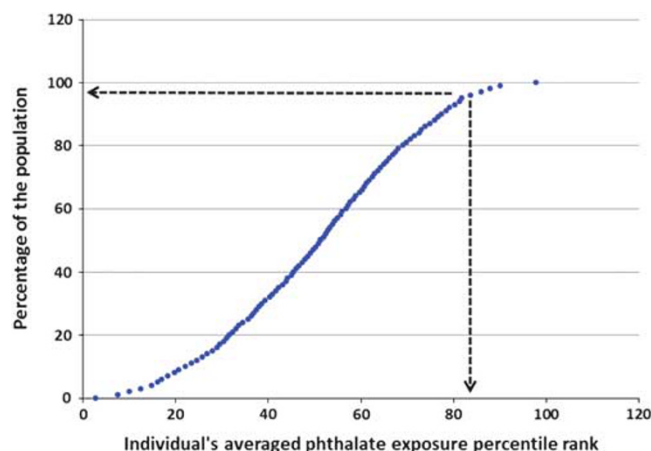


Figure 4. Cumulative distribution of individual's averaged phthalates exposure percentile rank. A cumulative distribution of the averaged exposure percentile ranks across the six phthalates was developed to explore what percentile rank of the individual phthalate exposure distributions, on average, would provide a summed coexposure estimate related to various percentile ranks of the summed exposure distribution for the NHANES data set.

the exposure estimates could further enhance the utility of bio-monitoring information. This could help in identifying key factors affecting the exposure and coexposure patterns of different substances, which could in turn enhance exposure prediction capabilities.

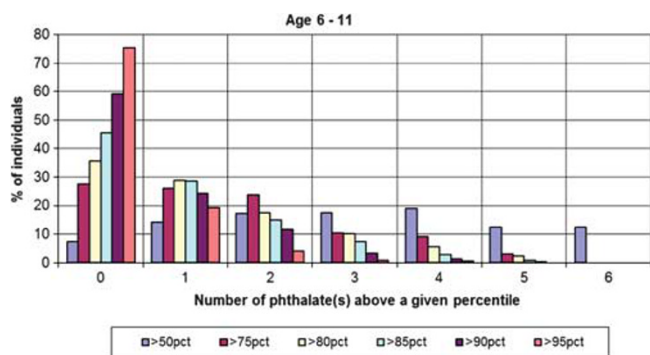


Figure 5. Percentage of children (6–11 years old) with the number of phthalates above a given percentile in the exposure sample distribution. Exposure distributions were calculated for each of the six phthalates examined for the study sample. The data were then tabulated to show the percentage of individuals within the study sample that had an estimated phthalate exposure greater than the given percentile of the exposure sample distribution.

For coexposures, assessment science is still emerging. RME and central-tendency exposure are typically calculated for single chemical exposure risk assessment. EPA³ advises that the RME at upper end of the range of risk estimates (generally between the 90th and 99.9th percentiles) should be selected in a probabilistic risk assessment for a single compound risk assessment. ECHA's guidance²⁶ on occupational exposure estimation recommends that the 90th percentile of an exposure distribution should be considered for the reasonable worst case as a general rule. For acute occupational exposure whose effects are transient and not severe, a higher percentile (e.g. 95th percentile) can be suggested as the reasonable worst case exposure estimator. Our analysis on NHANES phthalate data indicates that few individuals were exposed to multiple phthalates at 95th percentiles or greater, suggesting that the choice of 95th percentile value commonly used in single chemical risk assessment may be overly conservative for developing a realistic estimate of phthalate coexposure. For the data set evaluated, 95% of individuals had total exposure, with the averaged percentile rank at about 80 (Figure 4). However, caution must also be taken when applying this finding generally to other chemicals. The results will vary by chemicals and chemical sources. The use of biomonitoring data to determine coexposure patterns is of general applicability, but factors such as variation in use profiles, physical/chemical properties, and metabolic pathways can all contribute to coexposure patterns of specific chemicals. The analysis here is specific for phthalates. Similar analyses could be conducted to examine coexposure patterns for other chemicals.

Studies have shown that some subpopulations such as children, women, or non-Hispanic blacks usually had higher phthalate exposures than adolescents and adults, male, or Mexican Americans and non-Hispanic whites.^{17,23,24} While absolute values may differ, the analysis of this study demonstrated similar coexposure patterns for the six phthalates in the NHANES 2007/2008 total sample and subsample stratified by age, gender, and ethnicities (Figure 5 and Supplementary Figures 1 and 2).

CONCLUSION

So far, there is no simple approach that has been recommended to characterize coexposure patterns for multiple chemicals. This analysis identified that the NHANES data set can be a useful information source for an improved understanding of coexposures. Patterns of coexposure vary, suggesting that biomonitoring

data when available provide the best assessment of coexposure. The approach used in this analysis on coexposure patterns of the six phthalates can be extended to other chemicals as well.

This analysis focused solely on exposure patterns of these six phthalates. Patterns for contribution to coexposures are not equivalent to patterns of contribution to potential risk. An estimate of the potential risk from the phthalate coexposure would depend on the relative risk contribution from the individual phthalate, which was not addressed in this analysis.

The key to our analysis was the calculation of intake estimates from spot-urine samples represented in the 2007/2008 data set. However, given that spot-urine samples represent only a single moment in time, intake estimates are inherently variable, especially for phthalates for which dietary exposure is considered the likely route. Because the NHANES protocol for the 2007/2008 data set required participants in the morning session to begin a fast at 2300 hours the previous night, and participants in the afternoon and evening sessions to not fast, our intake estimates may be overestimated if it is assumed fasting time is an appropriate surrogate for time since last exposure.²⁷ Subsequently, the impact of spot-urine sampling methodology on intake estimates from phthalate exposures that are not predominately dietary is not fully understood.

For the total data set ($N = 2604$), the co-occurrence of multiple phthalates at the upper percentile of exposure was infrequent. There are no cases where all phthalates examined had exposure values > 95th percentile of each individual phthalates' exposure distribution. In only three individuals (0.12%), five phthalates fell above the 95th percentiles. For 75% of individuals, none of the six phthalates were above the 95th percentile of their respective exposure distributions. Coexposure patterns were similar regardless of subpopulations stratified by age, gender, and race. For the phthalates identified in this analysis, upper percentiles of total exposure tended to be dominated by a single ester, whereas exposures around the median of total exposure showed greater relative contribution of multiple substances.

CONFLICT OF INTEREST

All authors are employed by ExxonMobil, a commercial producer of DINP and DIDP.

REFERENCES

- USEPA. Risk Assessment Guidance for Superfund (RAGS): Volume I. Human Health Evaluation Manual (Part A), 1989; available at http://www.epa.gov/oswer/riskassessment/ragsa/pdf/rags_a.pdf.
- USEPA. *Supplemental Guidance to RAGS: Calculating the Concentration Term*. Environmental Protection Agency: Washington, DC, USA, 1992.
- USEPA. Risk Assessment Guidance for Superfund (RAGS): Volume III — Part A, Process for Conducting Probabilistic Risk Assessment 2001, available at http://www.epa.gov/oswer/riskassessment/rags3adt/pdf/rags3adt_complete.pdf.
- Meek M, Boobis A, Crofton K, Heinemeyer G, Van Raaij M, Vickers C. Risk assessment of combined exposure to multiple chemicals: a WHO/IPCS framework. *Regul Toxicol Pharmacol* 2011; **60**(Suppl 2): S1–S14.
- OECD OfEC-oad. WHO OECD ILSI/HESI International Workshop on Risk Assessment of Combined Exposures to Multiple Chemicals Workshop Report, 2011; available at [http://search.oecd.org/officialdocuments/displaydocumentpdf/?cote=env/jm/mono\(2011\)10&doclanguage=en](http://search.oecd.org/officialdocuments/displaydocumentpdf/?cote=env/jm/mono(2011)10&doclanguage=en).
- Interdepartmental Group on Health Risks from Chemicals (IGHRC). *Chemical Mixtures: A Framework For Assessing Risks (CR14)*. Institute of Environment and Health, Cranfield University: Cranfield, UK 2009.
- World Health Organization. *Harmonization Project Document 7; Assessment of Combined Exposures to Multiple Chemicals: Report of a WHO/IPCS International Workshop on Aggregate/Cumulative Risk Assessment*. WHO: Geneva, Switzerland, 2009; available at <http://www.oecd.org/officialdocuments/displaydocumentpdf/?cote=env/jm/mono%282011%2910&doclanguage=en>.
- Scientific Committees Scientific Committee on Consumer Safety (SCCS) Scientific Committee on Health and Environmental Risks (SCHER) Scientific Committee on Emerging and Newly Identified Health Risks (SCENIHR). Toxicity and Assessment of Chemical Mixtures (Preliminary Opinion approved for Public Consultation): European Commission, DG Health & Consumers 2011.

- 9 Food Quality Protection Act. United States Public Law 104-170, 1996; available at <http://epagov/pesticides/regulating/laws/fqpa/gpogatepdf>.
- 10 Schettler T. Human exposure to phthalates via consumer products. *Int J Androl* 2006; **29**: 134–139.
- 11 ATSDR. *Toxicological Profile for Di-n-Butyl Phthalate*. US Department of Health and Human Services, Public Health Service, Agency for Toxic Substances and Disease Registry: Atlanta, GA, USA 2001, available at <http://www.atsdr.cdc.gov/toxprofiles/tp135.pdf>.
- 12 David RM. Exposure to phthalate esters. *Environ Health Perspect* 2000; **108**: A440.
- 13 Kohn MC, Parham F, Masten SA, Portier CJ, Shelby MD, Brock JW *et al*. Human exposure estimates for phthalates. *Environ Health Perspect* 2000; **108**: A440.
- 14 Tietz N. In: Carl AB, Edward RA, David EB (eds). *Tietz Textbook Of Clinical Chemistry and Molecular Diagnostics*. Elsevier Saunders: St. Louis, MO, USA, 2006.
- 15 Remer T, Neubert A, Maser-Gluth C. Anthropometry-based reference values for 24-h urinary creatinine excretion during growth and their use in endocrine and nutritional research. *Am J Clin Nutr* 2002; **75**: 561–569.
- 16 Preau Jr JL, Wong L-Y, Silva MJ, Needham LL, Calafat AM. Variability over 1 week in the urinary concentrations of metabolites of diethyl phthalate and di (2-ethylhexyl) phthalate among eight adults: an observational study. *Environ Health Perspect* 2010; **118**: 1748.
- 17 Silva MJ, Barr DB, Reidy JA, Malek NA, Hodge CC, Caudill SP *et al*. Urinary levels of seven phthalate metabolites in the US population from the National Health and Nutrition Examination Survey (NHANES) 1999–2000. *Environ Health Perspect* 2004; **112**: 331.
- 18 Crépet A, Tressou J. Bayesian nonparametric model with clustering individual co-exposure to pesticides found in the French diet. *Bayesian Anal* 2011; **6**: 127–144.
- 19 Price PS, Han X. Maximum cumulative ratio (MCR) as a tool for assessing the value of performing a cumulative risk assessment. *Int J Environ Res Public Health* 2011; **8**: 2212–2225.
- 20 Tornero-Velez R, Egeghy PP, Cohen Hubal EA. Biogeographical analysis of chemical co-occurrence data to identify priorities for mixtures research. *Risk Anal* 2012; **32**: 224–236.
- 21 Kransler KM, Bachman AN, McKee RH. A comprehensive review of intake estimates of di-isononyl phthalate (DINP) based on indirect exposure models and urinary biomonitoring data. *Regul Toxicol Pharmacol* 2012; **62**: 248–256.
- 22 Kransler KM, Bachman AN, McKee RH. Estimates of daily di-isodecyl phthalate (DIDP) intake calculated from urinary biomonitoring data. *Regul Toxicol Pharmacol* 2013; **65**: 29–33.
- 23 Wittassek M, Angerer J. Phthalates: metabolism and exposure. *Int J Androl* 2008; **31**: 131–138.
- 24 Wittassek M, Koch HM, Angerer J, Brüning T. Assessing exposure to phthalates — the human biomonitoring approach. *Mol Nutr Food Res* 2011; **55**: 7–31.
- 25 Wittassek M, Wiesmüller GA, Koch HM, Eckard R, Dobler L, Müller J *et al*. Internal phthalate exposure over the last two decades—a retrospective human biomonitoring study. *Int J Hyg Environ Health* 2007; **210**: 319.
- 26 ECHA. Guidance on Information Requirements and Chemical Safety Assessment. Chapter R.14: Occupational Exposure Estimation, Ver2, 2012; available at http://echa.europa.eu/documents/10162/13632/information_requirements_r14_en.pdf.
- 27 Aylward LL, Lorber M, Hays SM. Urinary DEHP metabolites and fasting time in NHANES. *J Expos Sci Environ Epidemiol* 2011; **21**: 615–624.
- 28 Anderson W, Castle L, Scotter M, Massey R, Springall C. A biomarker approach to measuring human dietary exposure to certain phthalate diesters. *Food Addit Contam* 2001; **18**: 1068–1074.
- 29 Koch HM, Bolt HM, Preuss R, Angerer J. New metabolites of di (2-ethylhexyl) phthalate (DEHP) in human urine and serum after single oral doses of deuterium-labelled DEHP. *Arch Toxicol* 2005; **79**: 367–376.
- 30 Anderson WA, Castle L, Hird S, Jeffery J, Scotter MJ. A twenty-volunteer study using deuterium labelling to determine the kinetics and fractional excretion of primary and secondary urinary metabolites of di-2-ethylhexylphthalate and di-iso-nonylphthalate. *Food Chem Toxicol* 2011; **49**: 2022–2029.
- 31 Koch HM, Angerer J. Di-iso-nonylphthalate (DINP) metabolites in human urine after a single oral dose of deuterium-labelled DINP. *Int J Hyg Environ Health* 2007; **210**: 9–19.



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