Population-based estimates of environmental exposures using biomarkers can be difficult to obtain for a variety of reasons, including problems with limits of detection, undersampling of key strata, time between exposure and sampling, variation across individuals, variation within individuals, and the ability to find and interpret a given biomarker. In this article, we apply statistical likelihoods, weighted sampling, and regression methods for censored data to the analysis of biomarker data. Urinary metabolites for seven phthalates, reported by Blount et al., are analyzed using these methods. In the case of the phthalates data, we assumed the underlying model to be a log-normal distribution with the mean of the distribution defined as a function of a number of demographic variables that might affect phthalate levels in individuals. Included as demographic variables were age, sex, ethnicity, residency, family income, and education level. We conducted two analyses: an unweighted analysis where phthalate distributions were estimated with changes in the means of these distributions as a function of demographic variables, and a weighted prediction for the general population in which weights were assigned for a subset of the population depending on the frequency of their demographic variables in the general U.S. population. We used statistical tests to determine whether any of the demographic variables affected mean phthalate levels. Individuals with only a high school education had higher levels of di-n-butyl phthalate than individuals with education beyond high school. Subjects who had family income less than $1,500 in the month before sampling and/or only high school education had higher levels of n-butyl benzyl phthalate levels than other groupings. Di(2-ethylhexyl) phthalate was higher in males and/or in urban populations and/or in people who had family income less than $1,500 per month. Our findings suggest that there may be significant demographic variations in exposure and/or metabolism of phthalates and that health-risk assessments for phthalate exposure in humans should consider different potential risk groups. Key words: demographic factors, phthalates, risk assessment. Environ Health Perspect 110:405–410 (2002). [Online 11 March 2002] http://ehpnet1.niehs.nih.gov/docs/2002/110p405-410koo/abstract.html

Blount et al. (2) reported the concentrations of seven phthalate monoesters [monooctyl phthalate (MEP), monobutyl phthalate (MBP), monobenzyl phthalate (MBzP), monocyclohexyl phthalate (MCHP), mono-2-ethylhexyl phthalate (MEHP), monooctyl phthalate (MOAP), monoisononyl phthalate (MINP)] in the urine of 289 people, providing the first systematic compilation of data that address phthalate exposures to the general population from commercially important phthalate diesters. Kohn et al. (3) applied a simple pharmacokinetic model to estimate the total daily intake of phthalates that would result in the reported urinary concentrations of monoester metabolites. These intake estimates were used as a measure of total exposure to diethyl phthalate (DEP), di-n-butyl phthalate (DBP), n-butyl benzyl phthalate (BBP), diclohexyl phthalate (DCHP), di-(2-ethylhexyl) phthalate (DEHP), di-2-ethylhexyl phthalate (DEHP), di-n-octyl phthalate (DOP), di-n-nonyl phthalate (DNP).

Blount et al. (2) reported a considerable number of observations in which the analyte levels in urine were below the limit of detection (LOD) for the procedure being used. This analysis excluded analytes for which more than 25% of the studied individuals were below the LOD and discarded individuals below the LOD for analytes they did analyze. This represents a substantial loss of information. Maximum likelihood methods for censored observations (4-7) have been used for many years to analyze survival data and data for which some observations cannot be seen, but it is known that the observation is beyond some critical point. For urinary metabolite data, an observation below the LOD can be assumed to have a metabolite concentration less than the LOD. Methods have been developed for analyzing biomarkers of exposure—including observations below the LOD—by using statistical likelihoods and regression methods for censored data (8). Using a likelihood for censored data, these fractional pieces of information contribute to the overall interpretation of the data and can be used in a natural framework to estimate parameters and test for population differences. To account for strata differences of demographic factors, we estimated population-based exposures to phthalates using a weighted analysis in which weights were assigned for each individual group depending on the frequency of their demographic variables in the general U.S. population.

The aim of this study was to present methods for the analysis of exposure estimates based on urinary biomarker data accounting for strata differences and problems with LOD and to investigate the association between biomarker-based exposure estimates for phthalates and demographic factors in a human reference population.

Materials and Methods

Phthalate data. The data for this study were collected from adults during 1988–1994 as part of the Third National Health and Nutritional Examination Survey (NHANES III) (9). NHANES III analyzed urine...
samples from 289 subjects for 7 phthalate monoesters (MEP, MBP, MBEp, MCHP, MEHP, MOP, MINP). Urine samples were collected at different times throughout the day and were not first-morning voids. This sampling of the NHANES III population was not designed to be representative of the U.S. population but rather to serve as a reference range for a demographically described group. The population studied comprised noninstitutionalized adults ages 20–60 years (mean ± SD, 37.4 ± 10.6 years). The sex distribution (56% female) was similar across age groups. Racial distribution was weighted toward minority groups (Caucasian, 39%; African American, 30%; Mexican American, 23%; and other, 8%). The residency distribution was urban 57%, rural 43%. The family income was categorized by two groups (≥ $1,500 in the month before sampling, 56%; < $1,500 in the month before sampling, 44%), and the education level was categorized by two groups (high school diploma or less, 69%; education beyond high school, 31%).

Estimating intake levels from urinary metabolites. Kohn et al. (3) calculated the intake for each individual in the reference population as follows: The daily exposure can be estimated by using a linear two-compartment model. The normalized integrated rate equations are

\[ FE = \frac{1}{\mu_{\text{total}}} - \exp\left(-k_{\text{FUT}}t\right) \]  

\[ FU = \frac{k_{\text{FUT}}}{k_{\text{FUT}}} \left[1 - \exp\left(-k_{\text{FUT}}t\right)\right], \]

where \( FE \) and \( FU \) are the total and urinary fractions of the dose eliminated in time \( t \), and \( k_{\text{FUT}} \) are the apparent first-order rate constants for total elimination and elimination of urinary monoester, respectively. We calculated the two rate constants from the excreted fractions observed during the 24 hr after a single oral dose of diester, using Equations 1 and 2.

Assuming steady-state intake and metabolic clearance of the diester, the internal exposure rate for an individual was approximated by Equation 3 to be:

\[ \text{intake (μg/kg/day)} = \frac{ME(μg/g) \times CE(μg/kg/day)}{f \times 1,000(μg/g)} \times \frac{MW_{\text{d}}}{MW_{\text{m}}}. \]

where \( ME \) is the urinary concentration of monoester per g creatinine, \( CE \) is the excretion rate normalized by body weight, \( f \) is the ratio of urinary excretion to total elimination \( k_{\text{FUT}}/k_{\text{total}} \), and \( MW_{\text{d}} \) and \( MW_{\text{m}} \) are the molecular weights of the di- and monoesters, respectively.

Table 1 shows total fractional excretion (FE) and fractional urinary excretion of monoester (FU) during 24 hr after a single oral dose of diester.

**Table 1.** Total fractional excretion (FE) and fractional urinary excretion of monoester (FU) during 24 hr after a single oral dose of diester.

| Monoester | Diester | FE | FU |
|-----------|---------|----|----|
| Ethyl (MEP) | Diethyl (DEP) | 0.94* | 0.52* |
| n-Butyl (MBP) | Di-n-butyl (DBP) | 0.94 | 0.52 (10–17) |
| Benzyl (MBzP) | n-Butyl benzyl (BBP) | 0.70 | 0.36 (12–14) |
| Cyclohexyl (MCP) | Dicyclohexyl (DCHP) | 0.65 | 0.069 |
| 2-Ethylhexyl (MEHP) | Di(2-ethylhexyl) (DEHP) | 0.65 | 0.069 (15–19) |
| n-Octyl (MOP) | Di-n-octyl (DOP) | 0.65 | 0.043 (17) |
| i-Nonyl (MINP) | Di-i-nonyl (DINP) | 0.65* | 0.069* |

*Assumed to be the same as di-n-butyl phthalate. *Assumed to be the same as di(2-ethylhexyl) phthalate.

Statistical methods. Linear models are a common means of analyzing data to detect statistically significant differences between groups or for significant trends in the data as a function of some continuous variable. Let \( X \) denote the random variable associated with the daily intake level calculated using Equation 3 in a given individuals. For the analysis presented here, we assume that

\[ Y = \ln(X) = f_0(\theta) + \varepsilon, \]

where \( \ln \) denotes the natural log of \( X, f_0(\theta) \) is a function of a set of covariates denoted \( \theta \), and, depending on a set of parameters, \( \mu \), and \( \varepsilon \) is a random variable for which

\[ \varepsilon \sim N(0, \sigma^2). \]

That is, \( \varepsilon \) is normally distributed with mean 0 and variance \( \sigma^2 \). From Equation 4, it follows that

\[ E[Y] = f_0(\theta). \]

In the analysis that follows, we use a linear model to analyze the impact of age, sex, ethnicity, residence, family income, and education level to phthalate exposure data. The parameters are estimated by maximum likelihood in LIFEREG procedure in SAS 8.0 for Windows (SAS Institute, Cary, NC) to test for significant differences in mean phthalate levels as a function of age, sex, ethnicity, residence, family income, and education level to phthalate exposure data. The frequency of their demographic variables was of the form

\[ f_0(\theta) = \frac{1}{\sigma^2} \int_{-\infty}^{\infty} \Phi(z; \theta) N(z; \mu, \sigma^2) dz, \]

where \( n \) is the sample size (\( n = 289 \) for the phthalates example), \( \Phi(z; \mu, \sigma^2) \) is the density function for the normal distribution with mean \( \mu \) and variance \( \sigma^2 \) evaluated at \( z \), \( N(z; \mu, \sigma^2) \) is the cumulative density function for the normal distribution with mean \( \mu \) and variance \( \sigma^2 \), \( I_{\text{LOD}} \) is an indicator function such that \( I_{\text{LOD}} = 1 \) if the observed urinary metabolite can be quantified, and \( I_{\text{LOD}} = 0 \) if it is below the limit of detection. \( Y \) is the log of the daily intake level for the \( \theta \)th individual and \( LOD \) is the log of the limit of detection for the \( \theta \)th individual calculated as daily intake level.

We performed the regression analysis using the LIFEREG procedure in SAS 8.0 for Windows (SAS Institute, Cary, NC) to test for significant differences in mean phthalate levels as a function of age, sex, ethnicity, residency, family income, and education level to phthalate exposure data. The parameters are estimated by maximum likelihood in LIFEREG procedure, and probability density function is used if the observed urinary metabolite can be quantified, or cumulative density function is used if it is below the limit of detection (8). We conducted this analysis using an unweighted analysis where phthalate distributions were estimated with changes in the means of these distribution as a function of demographic variables. We estimate phthalates exposures using a weighted prediction for the general population in which weights were assigned for subset of the population depending on the frequency of their demographic variables in the general U.S. population. In this analysis, a composite distribution is formed by resampling from individual distributions for each significant demographic variable.

Results

As an initial step, we calculated correlations across phthalates and demographic factors. Within exposure estimates for phthalates,
DBP was highly correlated with BBP ($r = 0.52, p < 0.01$). We noticed relatively low correlation between BBP and DEHP ($r = 0.28, p < 0.01$). DBP showed a slight correlation with DCHP ($r = 0.13, p = 0.02$), DEHP ($r = 0.19, p < 0.01$), and DOP ($r = 0.13, p = 0.02$); BBP with DCHP ($r = 0.12, p = 0.02$) and DOP ($r = 0.13, p = 0.01$); and DCHP with DEHP ($r = 0.11, p = 0.04$), DOP ($r = -0.14, p = 0.01$), and DINP ($r = 0.19, p < 0.01$). Also, we noticed a slight correlation between DEP and DBP ($r = 0.18$) and between DOP and DINP ($r = 0.15$). BBP showed slightly significant correlation coefficients with age ($r = -0.11, p = 0.04$), family income ($r = 0.17, p < 0.01$), and education level ($r = -0.16, p < 0.01$); DCHP with education level ($r = 0.12, p = 0.03$); DEHP with sex ($r = -0.13, p = 0.02$), Mexican ethnicity ($r = 0.11, p = 0.04$), residency ($r = -0.13, p = 0.01$), and family income ($r = 0.14, p = 0.01$). There was a similar magnitude of correlation between DINP and education level ($r = -0.12, p = 0.04$), and DBP and education level ($r = -0.13, p = 0.02$). The reference values are male in sex, non-black in black ethnicity, non-Mexican in Mexican ethnicity, urban in residency, more than $1,500 in the month before sampling in family income, and high school diploma or less in education level.

Table 2 shows the results of the regression analysis using maximum likelihood methods as described in "Materials and Methods." Individuals with only a high school education had higher levels of DBP than individuals with education beyond high school ($p = 0.05$). Subjects who had family income less than $1,500 in the month before sampling and/or only high school education had higher levels of BBP than other groupings ($p < 0.05$). DEHP was higher in males and/or in urban populations and/or in people who had family income less than $1,500 per month ($p < 0.05$).

Figure 1 demonstrates the differences between estimates of DBP in subjects who had only a high school education or less versus subjects with education beyond high school. The fitted normal curve of log DBP in subjects who had a high school education or less was significantly shifted to right ($p = 0.02$). There were no censored observations in these samples, and the results indicate fairly close agreement between the observed data and the fitted normal distribution. Figure 2 illustrates the fit of the normal curve of log DEHP by residency; here the distribution was significantly shifted to the right in subjects who lived in the urban areas ($p = 0.01$). The proportion below the LOD is less in urban (17.7%) than in rural (27.2%), and both graphs show a considerable difference between the observed histogram and the plotted density due to the data points below the LOD.

Table 3 shows estimated phthalates exposure weighted using demographic characteristics in the general U.S. population. The mean of estimated exposure is 10.1 µg/kg/day for DEP, 1.66 µg/kg/day for DBP, 0.84 µg/kg/day for BBP, 1.26 × 10⁻⁵ µg/kg/day for DCHP, 0.41 µg/kg/day for DEHP, 6.16 × 10⁻⁵ µg/kg/day for DOP, and 8.99 × 10⁻⁷ µg/kg/day for DINP.

Discussion

Population-based estimates of environmental exposures using biomarkers can be difficult to obtain for a variety of reasons, including problems with limit of detection, under-sampling of key strata, time between exposure and sampling, variation across individuals, variation within individuals, and the ability to find and interpret a given biomarker. In this article, we present methods for analyzing biomarkers of exposure using statistical likelihoods, weighted sampling, and regression methods for censored data. Determination of normal ranges using biomonitoring data where measurements are below the LOD is a frequently encountered problem. Data sets in which concentrations below a fixed value are undetectable usually fit a normal or log normal distribution, and more adequate statistical methods can be used to determine their normal range. Maximum-likelihood estimation is a more appropriate statistical

Table 3 shows estimated phthalates exposure weighted using demographic characteristics in the general U.S. population.

![Figure 1](image1.png)

**Figure 1.** The fit of the normal curve to log DBP data by education; the means of these two distributions are significantly different ($p = 0.02$). Data were analyzed using a linear regression model with maximum likelihood methods that account for censoring; see equations in text.
method for the determination of normal range from left-censored data (4–7). Any statistical analysis depends on the assumption that the data can reasonably be regarded as a random sample from some underlying distribution. For the present case, data sets are available that are not left-censored. These data sets can be used to suggest suitable distributions for the censored samples, and techniques for estimation of parameters from such samples are straightforward. The log-normal distribution adequately fit these data (Figure 1) and were used for all of the biomarkers. In the case of our left-censored data, we used the corresponding cumulative probability distributions so that the likelihood functions for models involving censored data can easily be constructed and maximized.

Phthalates are used in the manufacture of a wide range of plastic and nonplastic products. Most of a phthalate dose is cleared in 24 hr and completely eliminated in 3–5 days (13,19–22). Because phthalates are lipophilic (23), it might be predicted that these compounds would accumulate in fat. However, with other lipophilic compounds, such as polychlorinated biphenyls, deposition of the compound into fat may not occur until several hours or several months after dosing (24–26). Because of the rapid metabolism of phthalates to more polar metabolites, these compounds are not sequestered in fat. Phthalates are widely distributed in the body, with the liver being the major, initial repository organ. Clearance from the body is rapid, and there is only a slight cumulative potential (16). Even though there is only a slight cumulative potential, phthalates are found in a wide variety of extensively used products, have been identified in all environmental compartments, and are a serious concern for the possibility of adverse effects. The acute toxicity of phthalates is low, with LD50 values ranging from 0.7 to > 20 g/kg (27); however, changes in lipid metabolism (28–30), testicular atrophy (31,32), alterations in xenobiotic metabolism (33,34), liver peroxisome proliferation (35), and carcinogenicity (36,37) have been observed. Regarding reproductive and developmental effects, phthalates vary in potency, with DEHP being the most potent and DBP and BBP the least potent (38–43).

Another difficulty in estimating the environmental hazard posed by phthalates is the lack of sufficient data documenting the human and wildlife exposure. Furtmann (46) has suggested that the main source of phthalates is consumer products, and that as a result of disposal of these products, there are considerable phthalate emissions into the environment. The estimated total loss to the environment of phthalates in Western Europe has been estimated as 7,740 tons/annum, or approximately 1% of total consumption (47). However, the use of such data in the assessment of environmental hazards for individual chemicals is problematic because the data are generalized, and estimates refer to total phthalates. Other more rigorous deterministic approaches based on measured or estimated levels in environmental media (food, soil, water, air) and human activity/consumption patterns have been used for estimating individual phthalate exposure (48). Recently, the intake of several phthalates was estimated from measured individual urinary phthalate by Kohn et al. (3) and were found to agree quite well with previous deterministic exposure estimates (3). Kohn et al. described in detail how the different metabolites can be derived from common precursor compounds or can arise from different parent compounds. For MBzP, the presumed parent compound is BBP; however, MBP has two parent compounds, BBP and DBP. For the other monoesters, the presumed parent compound is the diester with two of the same substituents as in the monoester. Estimates of exposure from biomarker data are based on real, not potential, dose, provide information on individual variation in exposure, and allow for a more rigorous evaluation of factors contributing to exposure. In this study, we investigated the association between biomarker-based exposure estimates for phthalates and demographic factors in a human reference population.

Figure 2. The fit of the normal curve to log DEHP data by residency; the means of these two distributions are significantly different ($p < 0.01$). Data were analyzed using a linear regression model with maximum likelihood methods that account for censoring; see equations in text.

Table 3. Estimated phthalates exposure (µg/kg/day) weighted using demographic characteristics in the general US population and using regression parameters which are significant ($p < 0.15$) from the LIFEREG procedure.*

| Phthalates | Variablea | Mean | Median | 5th percentile | 95th percentile |
|-----------|-----------|------|--------|----------------|----------------|
| DEP       | Ethnicity (black) | 10.1 | 10.2   | 0.43           | 229            |
|           | Education, ethnicity (Mexican) | 1.66 | 1.66   | 0.31           | 8.78           |
| BBP       | Family income, education | 0.84 | 0.85   | 0.19           | 3.65           |
| DCHP      | Family income | 1.26 × 10^{-5} | 1.30 × 10^{-5} | 1.18 × 10^{-9} | 0.14 |
| DEHP      | Sex, residence, family income | 0.41 | 0.41   | 0.015          | 11.3           |
| DOP       | Residence, education | 6.16 × 10^{-5} | 6.26 × 10^{-5} | 2.19 × 10^{-9} | 1.56 |
| DINP      | None | 8.99 × 10^{-7} | 9.28 × 10^{-7} | 4.25 × 10^{-13} | 1.67 |

aData from U.S. Census Bureau (18). bBelow 0.15 significant level.
especially with respect to potentially susceptible populations. Our analysis suggests that people with a high school education or less have higher urinary output of DBP and BBP metabolites; individuals with a family income less than $1,500 in the month before sampling have higher urinary output of BBP and DEHP metabolites; and males and urban populations have higher urinary output of DEHP metabolites. The analysis used assumed that the pharmacokinetics of these compounds is the same in all individuals; this may not be true because genetic polymorphisms in the genes controlling the metabolism and elimination of phthalates may exist and could have an impact on levels of these metabolites in the urine. Hence, our findings may derive from differences in actual exposures, differences in metabolism, or a combination of these. Further study is needed to determine which of these may drive the observed differences.

Our sampled data have strata differences in demographic factors compared with the general U.S. population. We found significant variables in the regression model using the LIFEREG procedure weighted toward the general U.S. population. This approach yielded estimated distributions with changes in the mean and fifth percentile of phthalate intake estimates using this approach, even for a small, nonrepresentative sample as demonstrated here can be a useful approach for evaluating human exposures, but should be repeated in future NHANES, leading to larger cumulative sample sizes to be used for deriving national estimates of both current exposure levels and exposure trends. The data available to date show that urinary levels of MEP, MBzP, MBP from NHANES 1999 are lower than those from NHANES III. Further efforts will focus on evaluating the association between biomarker-based exposure estimates for phthalates and demographic factors in this larger human reference population.

In summary, we developed methods for analyzing biomarkers of exposure using statistical likelihoods, weighted sampling, and regression methods for censored data and analyzed the association between biomarker-based exposure estimates for phthalates and demographic factors in a human reference population. Our findings suggest that there may be significant demographic variations in exposure and/or metabolism of phthalates, and that health-risk assessments for phthalate exposure in humans should consider different potential risk groups. These findings support and extend previous information on human phthalate exposure and should prove useful in accurately quantifying human risk of exposure to these compounds, identifying factors contributing to higher exposures and opportunities for reducing those exposures, and stimulating additional research on sources of exposure to phthalates.

REFERENCES AND NOTES
1. Harris CA, Henttu P, Parker MG, Sumpter JP. The estrogenic activity of phthalate esters in vitro. Environ Health Perspect 105:802–811 (1997).
2. Blount BC, Silva MJ, Caulfield SP, Needham LL, Pirkle JL, Sampson EJ, Lucier GW, Jackson RJ, Brock JW. Levels of seven urinary phthalate metabolites in a human reference population. Environ Health Perspect 108:979–982 (2000).
3. Kohn MC, Parham F, Masten SA, Portier CJ, Shelby MD, Brock JW, Needham LL. Human exposure estimates for phthalates. Environ Health Perspect 108:440–442 (2000).
4. Tsy TY, Chen IW, Maxow HT, Himmelfarb J, 1. A statistical method for determining normal ranges from laboratory data including values below the minimum detectable value. Clin Chem 25:2011–2014 (1979).
5. Vilcinskas IR, Marriott F. Evaluation of censored contamination data. Food Addit Contam 12:637–644 (1995).
6. Lindsey JK, Byrom WD, Wang J, Jarvis P, Jones B. Generalized nonlinear models for pharmacokinetic data. Biometrics 56:81–88 (2000).
7. Burmester DE, Wilson AM. Fitting second-order finite mixture models to data with many censored values using maximum likelihood estimation. Risk Anal 20:261–271 (2000).
8. Tohno J. Estimation of relationships for limited dependent variables. Econometrica 26:24–36 (1958).
9. Department of Health and Human Services. National Center for Health Statistics. Third National Health and Nutrition Examination Survey, 1988–1994, NHANES III Laboratory Data File. Public Use Data File Documentation No. 7620. Hyattsville, MD:Centers for Disease Control and Prevention, 1996.
10. Tanaka A, Matsumoto A, Yamada T. Biochemical studies on phthalic esters. III. Metabolism of dibutyl phthalate (DBP) in animals. Toxicology 1:919–122 (1978).
11. Foster PM, Cook MW, Toth LS, Walters DG, Gangoli SD. Differences in urinary metabolic profile from di-n-butyl phthalate-treated rats and hamsters. A possible explanation for species differences in susceptibility to testicular atrophy. Drug Metab Dispos 11:89–91 (1983).
12. Nasdellie C, Picard K, Valentin I, Lhuigend JC, Chagnon MC. Metabolism of n-butyl benzyl phthalate in the female Wistar rat. Identification of new metabolites. Food Chem Toxicol 37:905–917 (1999).
13. Eigenberg DA, Bozqian HP, Carter DE. Distribution, excretion, and metabolism of butylbenzyl phthalate in the rat. J Toxicol Environ Health 17:445–456 (1986).
14. Castle L. Personal communication (1992).
15. Peck CC, Albrow PW. Toxic potential of the plasticizer di(2-ethylhexyl) phthalate in the context of its disposition and metabolism in primates and man. Environ Health Perspect 45:11–17 (1982).
16. Klouwe WM. Overview of phthalate ester pharmacokinetics in mammalian species. Environ Health Perspect 45:9–19 (1982).
17. Albro PW, Moore B. Identification of the metabolites of simple phthalate diesters in rat urine. J Chromatogr 94:209–218 (1974).
18. U.S. Census Bureau. Statistical Abstract of the United States: 2000. Available: http://www.census.gov/statab/www/ [cited 1 August 2001].
19. Williams DT, Blanchfield BJ. The retention, distribution, excretion and metabolism of dibutyl-7-14C in the rat. J Agric Food Chem 23:854–858 (1975).
20. Tanaka A, Kadoji T, Takahashi T, Yamada T. Biochemical studies on phthalic esters. I. Elimination, distribution and metabolism of di(2-ethylhexyl) phthalate in rats. Toxicology 1975:253–264 (1975).
21. Daniel JW, Bratt H. The absorption, metabolism and tissue distribution of di(2-ethylhexyl) phthalate in rats. Toxicology 251:65–175 (1974).
22. Williams DT, Blanchfield BJ. The retention, excretion and metabolism of di(2-ethylhexyl) phthalate administered orally to the rat. Bull Environ Contam Toxicol 11:371–378 (1974).
23. Leyder F, Boulanger P. Ultraviolet absorption, aqueous solubility and octanol-water partition for several phthalates. Bull Environ Contam Toxicol 30:152–157 (1984).
24. Matthews HB, Anderson MW. The distribution and excretion of 2,4,5,2',5'-pentachlorobiphenyl in the rat. Drug Metab Dispos 3:211–219 (1975).
25. Pittman KA, Wiener M, Treble DH. Mirex kinetics in the rhesus monkey. I. Pharmacokinetic model. Drug Metab Dispos 3:289–295 (1975).
26. Rozman T, Rozman K, Williams J, Greim H. Enhanced fecal excretion of mirex in rhesus monkeys by 5% mineral oil in the diet. Drug Metab Dispos 4:251–252 (1981).
27. Adachi T, Takahashi T, Yamada T. Biochemical studies on phthalic esters: review of the literature. Environ Health Perspect 43–26 (1973).
28. Reddy JK, Moody DE, Azarnoff DL, Rao MS. Di-(2-ethyl...
hexyl) phthalate: an industrial plasticizer induces hypolipidemia and enhances hepatic catalase and carnitine acetyltransferase activities in rats and mice. Life Sci 18:941–946 (1976).

29. Bell FP, Patt CS, Brundage B, Gillies PJ, Phillips WA. Studies on lipid biosynthesis and cholesterol content of liver and serum lipoproteins in rats fed various phthalate esters. Lipids 13:66–74 (1978).

30. Oishi S, Hiraga K. Effects of monoesters of o-phthalic acid on serum lipid composition of rats. Toxicol Lett 14:79–84 (1982).

31. Creasy DM, Foster JR, Foster PMD. The morphological development of di-n-pentyl phthalate induced testicular atrophy in the rat. J Pathol 139:309–321 (1983).

32. Oishi S, Hiraga K. Testicular atrophy induced by di-2-ethylhexyl phthalate: effect of zinc supplement. Toxicol Appl Pharmacol 70:43–48 (1983).

33. Aitio A, Parkki M. Effect of phthalate esters on drug metabolizing enzyme activities in rat liver. Arch Int Pharmacodyn Ther 235:187–195 (1978).

34. Walseth F, Toftgard R, Nilsen OG. Phthalate esters I: Effects on cytochrome P-450 mediated metabolism in rat liver and lung, serum enzymatic activities and serum protein levels. Arch Toxicol 50:1–10 (1982).

35. Moody DE, Reddy JK. Hepatic peroxisome proliferators form a novel class of chemical carcinogens. Nature 283:397–398 (1980).

36. Heindel JJ, Powell CJ. Phthalate ester effects on rat Sertoli cell function in vitro: effects of phthalate side chain and age of animal. Toxicol Appl Pharmacol 115:116–123 (1992).

37. Gray LE Jr, Wolf C, Lambright C, Mann P, Price M, Cooper RL, Ostby J. Administration of potentially antiandrogenic pesticides (procymidone, linuron, iprodione, chlorsulfuron, p,p’-DDE, and ketoconazole) and toxic substances (diisobutyl- and diethylhexyl phthalate, PCB 169, and ethane dimethane sulphonate) during sexual differentiation produces diverse profiles of reproductive malformations in the male rat. Toxicol Ind Health 15:94–118 (1999).

38. Kluwe WM, McConnell EE, Huff JE, Haseman JK, Douglas JF, Hartwell VW. Carcinogenicity testing of phthalate esters and related compounds by the National Toxicological Program and the National Cancer Institute. Environ Health Perspect 45:129–133 (1982).

39. Ema M, Itami T, Kawasaki H. Teratogenic phase specificity of butyl benzyl phthalate in rats. Toxicology 79:11–19 (1993).

40. Foster PM, Thomas LV, Cook MW, Gangoli SD. Study of the testicular effects and changes in zinc excretion produced by some n-alkyl phthalates in the rat. Toxicol Appl Pharmacol 54:392–398 (1980).

41. Kluwe WM, McConnell EE, Huff JE, Haseman JK, Douglas JF, Hartwell VW. Carcinogenicity testing of phthalate esters and related compounds by the National Toxicological Program and the National Cancer Institute. Environ Health Perspect 45:129–133 (1982).

42. Heindel JJ, Powell CJ. Phthalate ester effects on rat Sertoli cell function in vitro: effects of phthalate side chain and age of animal. Toxicol Appl Pharmacol 115:116–123 (1992).

43. Gray LE Jr, Wolf C, Lambright C, Mann P, Price M, Cooper RL, Ostby J. Administration of potentially antiandrogenic pesticides (procymidone, linuron, iprodione, chlorsulfuron, p,p’-DDE, and ketoconazole) and toxic substances (diisobutyl- and diethylhexyl phthalate, PCB 169, and ethane dimethane sulphonate) during sexual differentiation produces diverse profiles of reproductive malformations in the male rat. Toxicol Ind Health 15:94–118 (1999).

44. Pirkle JL, Sampson EJ, Needham LL, Patterson DG, Ashley DL. Using biological monitoring to assess human exposure to priority toxicants. Environ Health Perspect 103(suppl 3):45–48 (1995).

45. Furtmann RNV. Phthalates in the aquatic environment. Report no. 6/93. Brussels:European Chemical Industry Council (European Council for Plasticisers and Intermediates, 1996).

46. ECPI. Phthalate Esters Used in PVC. Assessment of the Release, Occurrence and Possible Effects of Plasticizers in the Environment [Partial Copy]. Brussels:European Chemical Industry Council (European Council for Plasticisers and Intermediates, 1996).

47. The National Toxicology Program (NTP) Center for the Evaluation of Risk to Human Reproduction (CERHR). Available: http://cerhr.niehs.nih.gov [cited 1 August 2001].

48. CDC. National Report on Human Exposure to Environmental Chemicals. Available: http://www.cdc.gov/nceh/dls/report/default.htm [cited 1 October 2001].