

Oxidative stress and inflammatory markers in relation to urinary phthalate metabolites in NHANES



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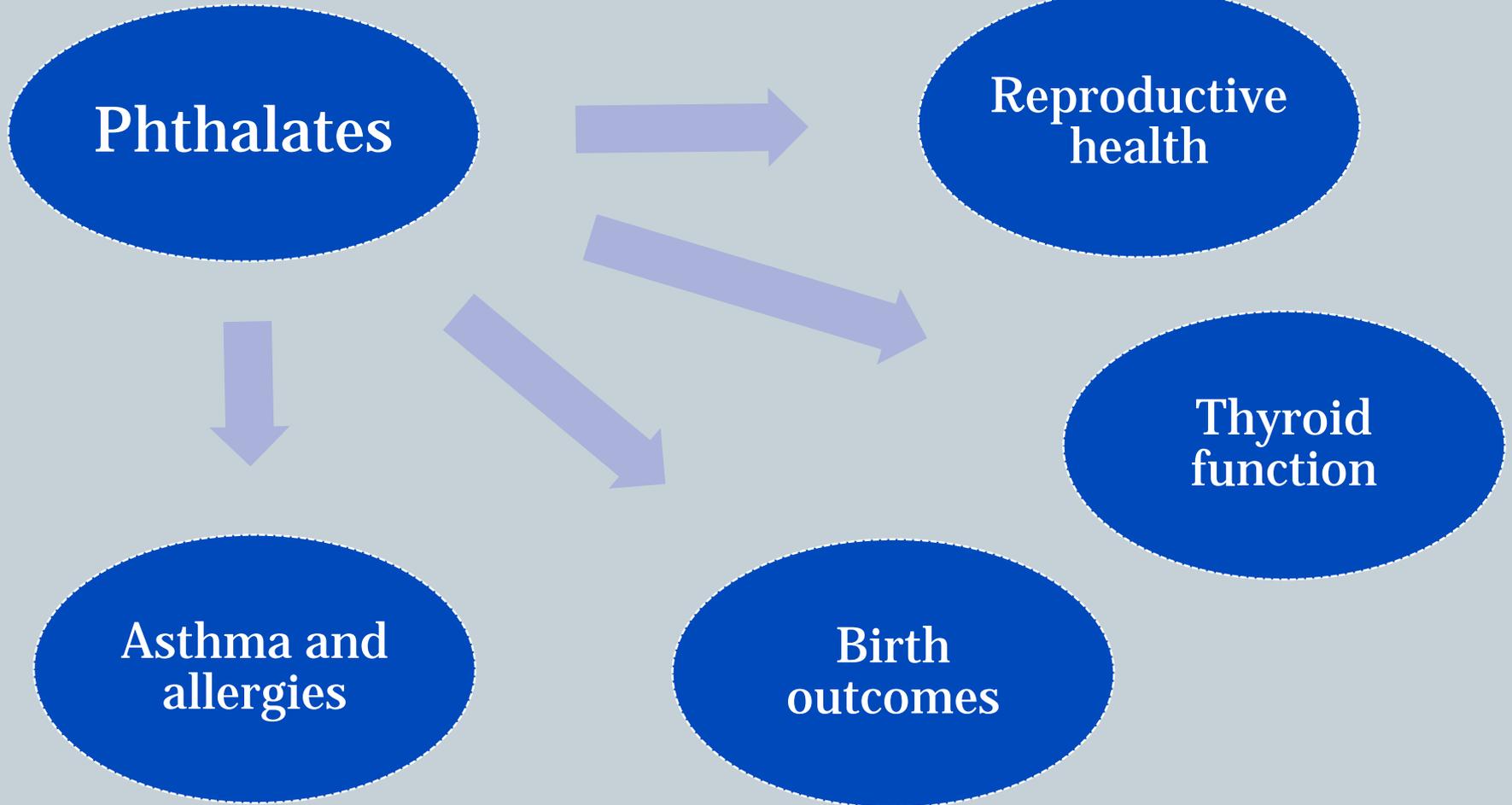


Environmental exposure to phthalates



- Phthalate production estimated at over 470 million tons per year (US EPA 2006)
 - High molecular weight (HMW) phthalates used as plasticizers in medical tubing, vinyl flooring, shower curtains, rain jackets, plastic bags etc.
 - Low molecular weight (LMW) phthalates used as solvents and in personal care products like soap, deodorants and lotions
- Exposure occurs primarily through ingestion of contaminated food and water (HMW), and secondarily through inhalation of contaminated air (LMW) (CDC 2011)

Phthalates are associated with multiple adverse health outcomes



Potential mediators of phthalate effects



- Oxidative stress and inflammation have been implicated as mechanistic intermediates in *in vitro* and animal studies
- Several recent epidemiologic studies have demonstrated associations with phthalate exposure in humans using various markers
- Here we examined relationships between phthalate metabolites and changes in systemic oxidative stress and inflammation, as indicated by particular biomarkers :
 - **Oxidative stress: Bilirubin**
 - **Inflammation: Alkaline phosphatase (ALP), absolute neutrophil count (ANC) and adjusted ferritin**



Methods



- **Study population: NHANES 1999-2006 (N=10,026)**
 - Spot urine samples taken for phthalate exposure measures
 - Blood serum or plasma used for assessing outcome levels
 - Covariates: Age, sex, race/ethnicity, poverty income ratio (PIR), serum cotinine and urinary creatinine
- **Statistical analysis**
 - Performed using SAS version 9.2 (SAS institute, Cary, NC)
 - Descriptive statistics
 - Multivariable regression models
 - Quintile analysis to examine non-linear relationships
 - Stratified analysis by age group, sex and race/ethnicity

Results: Descriptive statistics

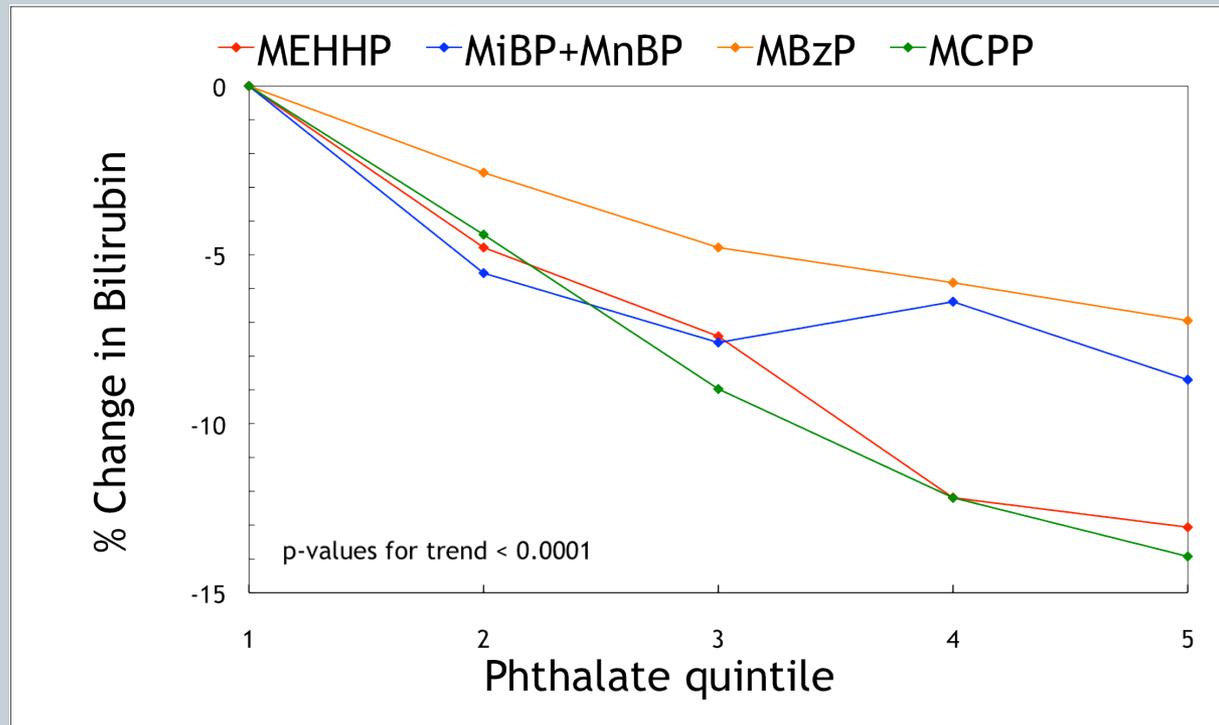


Parent compound	Metabolite	Years measured	N	% > LOD	Geometric mean	Median
DEHP	MEHP	1999-2006	10031	80.6	2.99	2.76
	MEHHP	2001-2006	7600	99.2	21.2	18.3
	MEOHP	2001-2006	7600	98.3	13.9	12.1
	MECPP	2003-2006	4930	99.9	35.0	30.0
DBP	MnBP	2001-2006	7600	99.1	18.9	18.2
	MiBP	2001-2006	7600	90.7	3.57	3.57
DEP	MEP	1999-2006	10026	99.9	167	145
BzBP	MBzP	1999-2006	10031	98.6	13.0	12.6
DOP	M CPP	2001-2006	7600	96.5	2.43	2.34
DiDP	MCNP	2005-2006	2417	91.5	2.67	2.48
DiNP	MCOP	2005-2006	2417	96.3	5.29	4.56

Results: Oxidative stress



- Bilirubin inversely associated with all phthalate metabolites except MCNP and MCOP



Results: Inflammation



Alkaline phosphatase

- Positively associated with MEHHP, MEOHP, MnBP, MiBP, DBPCOM, MBzP, MCPP and MCNP ($p < 0.01$)
- Inversely associated with MEHP and MEP

Absolute neutrophil count

- Positively associated with MEHHP, MEOHP, MnBP, MiBP, DBPCOM, MBzP and MCPP ($p < 0.01$)
- No significant inverse relationships

Adjusted ferritin

- Positively associated with MEHP, MEHHP and MEOHP ($p < 0.05$)
- Inversely associated with MEP

Discussion: Oxidative stress



- **Reliability as a marker**
 - Decreased bilirubin highly correlated with 8-OHdG (Tsukahara et al. 2003; Yoshida et al. 2001)
 - At lower levels correlated with MDA (Shekeeb et al. 2008)
 - In the present study, only slight correlated with gammaglutamyltransferase (GGT), and in the opposite direction expected
- **Evidence for a phthalate relationship—past and present**
 - Past studies have indicated MEHP, MEHHP and MEP are inducers of oxidative stress in humans, as they have been related to increased sperm DNA damage, urinary 8-OHdG and MDA and serum GGT (Hauser et al. 2007; Hong et al. 2009; Ferguson et al. 2011)
 - In the present study the same metabolites (except MEP) and several more are associated with bilirubin

Discussion: Inflammation



Alkaline phosphatase

- Positive correlation with C-reactive protein (CRP) (Webber et al. 2010)
- Consistent with previous results of positive association between CRP and MBzP and MiBP (Ferguson et al. 2011)
- Additional associations observed

Absolute neutrophil count

- Used in several studies as a marker of inflammation in response to environmental exposures (Frost-Pineda et al. 2011; Yang et al. 2009)
- Consistent with previous results and additional phthalate metabolites indicated to be associated with inflammation

Adjusted ferritin

- Acute phase protein indicative of inflammatory response, similar to CRP (Northrop-Clewes 2008)
- Slightly smaller sample size ($N \leq 5303$), but significant associations observed with all DEHP metabolites

Conclusions and future directions



- Phthalate metabolites MBzP, MCPP, and the metabolites of DEHP and DBP are associated with biomarkers indicative of increased inflammation and oxidative stress
- ALP, ANC and adjusted ferritin may be useful markers of inflammation, and bilirubin a useful marker of oxidative stress, in the study of health impacts of other low-dose environmental chemical exposures
- **Future directions:**
 - Use more robust study designs with longitudinal information and repeat urine samples for exposure assessment
 - Examine associations in sensitive subgroups
 - Explore other potential markers in large population-based studies

Thank you!



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