Biomonitoring of 21 phthalate metabolites in Slovak pre-school and school age children related to their consumer practices

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Abstract

Consumer practices are considered to be the main source of human exposure to phthalates. The aim of our study was biomonitoring of low molecular weight phthalate (LMWP) and high molecular weight phthalate (HMWP) metabolites in Slovak pre-school (n=100; age range 1-6 years) and school age children (n= 96; age range 7-15 years) and relate exposures to consumer practices. We used high performance liquid chromatography and tandem mass spectrometry for spot urine sample analysis to determine concentrations of 21 phthalate monoesters, metabolites of 11 LMWP and HMWP diesters. Median concentrations of ∑LMWP (177.32 $\mu g.L^{-1}$) were close to the ∑HMWP (169.83 $\mu g.L^{-1}$) metabolites. We observed significantly ($p \leq 0.05$) higher concentrations of 5 LMWP and 4 HMWP metabolites in school-age compared to preschool-age children. Data showed several statistically significant associations ($p \leq 0.05$) between concentrations of phthalate metabolites and the consumption of food and beverages. Interestingly, we observed for all HMWP metabolites (except mono(2-ethylhexyl) phthalate MEHP) and 1 LMWP metabolite (mono-methyl phthalate MMP) significantly lower concentrations in children who used at least 2 personal care products in comparison to non-users. Unexpected results suggest that potential routes of exposure differ from consumer practices included in our study.

Keywords: biomonitoring, phthalates, children, consumer practices

1. Introduction

Phthalates are man-made chemicals present in many consumer products. They can enter human body by ingestion, inhalation or dermal absorption (Beko et al., 2013). Children can be exposed most often by contaminated food (e.g. fat rich food- meat and dairy products) (Serrano et al., 2014), personal care products (e.g. shampoos, sunscreen and body creams) (Dodson et al., 2012) or plastic toys (Ashworth et al., 2018).

2. Material and Methods

2.1. Study design

The recruitment was done in day-care centres and schools during the years 2015-2016 among children in pre-school (n=100, age range 1-6 years) and school age (n= 96, age range 7-15 years) living in the Bratislavsky (n=50), Trnavsky (n=48), Nitriansky (n=70) and Presovsky (n=28) district of Slovakia. Parents signed an informed consent and completed a questionnaire for monitoring of sex, age and consumer practices of child during the last 48 hours before sampling (spot urine sample).

2.2. Chemical analysis

Spot urine samples were analysed for 21 phthalate metabolites [mono-methyl phthalate (MMP), mono-ethyl (MEP), mono-benzyl (MBzP), mono-iso-butyl (MiBP), 2OH-mono-iso-butyl (2OH-MiBP), mono-n-butyl (MnBP), 3OH-mono-n-butyl (3OH-MnBP), mono-cyclo-hexyl(MCHP), mono-n-pentyl(MnPeP), mono(2-ethylhexyl) (MEHP), mono(2-ethyl-5-hydroxyhexyl) (5OH-MEHP), mono(2-ethyl-5-oxohexyl) (5oxo-MEHP), mono-(2ethyl-5-carboxy pentyl) (5cx-MEPP), 7-OH-(mono-methyl-octyl) (OH-MiNP), 7-oxo-(mono-methyl-octyl) (oxo-MiNP), 7-carboxy-(mono-methyl-heptyl) (cx-MiNP), 6-OH-mono-propyl-heptyl (OH-MiDP), 6-oxo-mono-propyl-heptyl (oxo-MiDP), mono(2,7-methyl-7-carboxy-heptyl) (cx-MiDP), mono-n-octyl (MnOP) and mono-3-carboxy-propyl phthalate (MCPP)] at the Institute for Prevention and Occupational Medicine (IPA) laboratory via on-line HPLC–MS/MS using internal isotope-labelled standards according to method published previously elsewhere (Koch et al., 2013; 2017). Quality control material (pooled native urine) and blank samples were analyzed with each analytical batch. Limits of quantification (LOQ) for metabolites ranged between 0.2 - 1.0 $\mu g.L^{-1}$. Statistical analyses results were considered significant by $p \leq 0.05$.

3. Results and Discussion

At least 12 phthalate metabolites in concentrations above LOQ were detected in 100% of samples. Significant differences in phthalate concentrations between genders were not identified. We found higher concentrations of 5 LMWP and 4 HMWP in school age compared to pre-school age children ($p\leq 0.044$). We observed significantly higher concentrations of MEP, MnBP and OH-MnBP in the Presovsky district ($p\leq 0.034$).

Compared to the DEMOCOPHES biomonitoring study (Den Hond et al., 2015) concentrations of MEP, MBzP and DEHP metabolites were lower in present study, while levels of MiBP and MnBP were equal or higher.

Table 1. Median phthalate concentrations ($\mu g.L^{-1}$)

LMWP*		HMWP*			
MMP	2.7	MEHP	2.1	OH-MiDP	1.4
MEP	19.8	OH-MEHP	18.4	oxo-MiDP	0.5
MBzP	2.7	oxo-MEHP	12.2	cx-MiDP	1.0
MnBP	50.1	cx-MEPP	25.3	∑DiDP[3]	2.9
OH-MnBP	6.6	∑DEHP[1]	59.3		
MiBP	43.4	OH-MiNP	7.2		
OH-MiBP	16.0	oxo-MiNP	2.6		
MCPP	1.8	cx-MiNP	7.7		
		∑DiNP[2]	18.6		
∑LMWP	177.3			∑HMWP	169.8

*MCHP, MnPeP, MnOP- more than 97% of samples ≤LOQ

[1]∑DEHP= MEHP + oxo-MEHP + OH-MEHP + cx-MEPP

[2]∑DiNP= OH-MiNP + oxo-MiNP + cx-MiNP

[3]∑DiDP= OH-MiDP + oxo-MiDP + cx-MiDP

In our study household usage of plastic food-boxes was associated with higher levels of OH MiBP ($p=0,049$) and eating of food stored in plastic box (during the last 48 hours) with higher levels of MEP (in girls, $p=0,025$). In case of food consumption, we detected higher concentrations of MiBP, OH-MiBP and MEHP ($p\leq 0.026$) in girls consuming packaged salami and MMP ($p= 0.042$) in boys consuming margarines, compared to non-consumers. Multiple regression analyses showed significantly lower concentrations of MMP in children who consumed only 2 or 3 of food products in comparison to consumers of 4 and 5. These results are in accordance to study Sakhi et al. (2014) which specify some of the top food groups (bread, meat and meat products, fats and oils, milk products) contributing to dietary exposure to phthalate diesters. Similar phthalate-containing food groups are indicated by Serrano et al. (2014). Inconsistently with mentioned studies we observed higher concentrations of MEP in children, who consumed ≤1 of food products compared to consumers of 2 or 3 ($p\leq 0.026$) and higher concentrations of HMWP metabolites ($p\leq 0.043$) and MEP ($p= 0.041$) in non-consumers of baguettes and milk products (respectively).Main sources of dermal exposure to phthalates are personal care products. Our results pointed on the perfumes (MCPP in boys, $p=0.029$), body lotions (MiBP, OH-MiBP, $p\leq 0.029$) and wet wipes use (MCPP, MnBP in girls, $p\leq 0.018$) as routes of exposure. These observations are in accordance to studies Cutanda et al., 2015; Koch et al., 2013 and Dodson et al., 2012. Interestingly, multiple regression analyses showed for all HMWP metabolites (except MEHP) ($p\leq 0.029$) and 1LMWP metabolite (MMP; $p= 0.016$) significantly lower concentrations in children who used at least 2 and more of personal care products compared to non-users.

4. Conclusion

In view of study results, we consider consumer practices an important route of children phthalate exposure, but we suppose some additional sources and routes of exposure too. Some of them could be indoor environment of daycares and schools (Beko et al., 2013) or some other types of consumer products, we did not ask about.

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