Update from CDC: Phthalates and Phthalate Alternatives

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Biomonitoring California Scientific Guidance Panel Meeting
July 16, 2015
Oakland, CA
Outline

- Phthalates exposure generalities
- Changes in exposures
  - Use of NHANES
  - Archived samples: DINCH example
  - German Environmental Specimen Bank
- Selection of phthalate biomarkers
  - DiNP
  - DBP and DiBP
- Toxicology vs exposures
  - DPP
- Future work
What are Phthalates?

- Widely used industrial chemicals
  - As plasticizers of PVC
    - Miscellaneous products
    - Medical devices, tubing and blood bags
  - In consumer & personal care products
    - Fragranced products, cosmetics
    - Paints, ink & lacquers
    - Medications

- Adverse health outcomes in experimental animals exposed to high doses of phthalates

- Emerging data on potential human effects at background exposure levels

- Metabolites as biomarkers of exposure
The Human Exposure Scenario

- Controlled conditions, as in animal studies, do not apply
- Numerous and even unknown exposure routes and sources
- Unknown dose, duration, frequency and timing
- People exposed to chemical "cocktails" (multiple/mixtures)
- Biomonitoring to assess exposures
CDC’s Phthalates Biomonitoring Program Areas

- Assess exposure to phthalates & alternatives
- Assess associations between exposure & health
- R&D to improve Biomonitoring practices
  - Develop analytical methods
  - Identify & validate biomarkers
    - Replacement chemicals
  - Develop Standard Reference Materials
- Capacity building
  - Public Health Laboratories: Performance testing
Biomonitoring Methods

General requirements
- Sensitive
- Specific/Selective
- Accurate
- Precise

Biomonitoring-specific
- Minimum sample volume
  - Reduce solvent use & waste
- Multianalyte & high-throughput
  - Increase efficiency
- Reproducible
- Include QA/QC program
  - Accountability
- Automated
  - Cost effective

Best compromise
Accuracy: The Importance of Quantification

- Analytical standards
  - Custom synthesis
- Analytical method
- Well-maintained instrumentation
- Trained personnel
- External Quality Assessment Programs
  - G-EQUAS ([http://www.g-equas.de/](http://www.g-equas.de/))
    - Four DEHP metabolites, MnBP, MiBP, MBzP
  - Accuracy of standards (neat vs solution)

Accuracy Investigation of Phthalate Metabolite Standards

Eric Langlois*, Alain LeBlanc, Yves Simard and Claude Thellen
Centre de Toxicologie du Québec (CTQ), Institut National de Santé Publique du Québec (INSPQ), Québec, Québec, G1V 5B3, Canada
NIST SRMs

- Urine from smokers (3672)
- Urine from non-smokers (3673)
- First frozen urine reference materials characterized for organic environmental contaminants
  - 11 phthalate metabolites

Table 2. Reference Mass Fraction Values for Selected Phthalate Metabolites in SRM 3672

<table>
<thead>
<tr>
<th>Phthalate Metabolites</th>
<th>Mass Fraction (µg/kg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mono-carboxyonyl phthalate isomers</td>
<td>1.92 ± 0.06</td>
</tr>
<tr>
<td>Mono-carboxyetyl phthalate isomers</td>
<td>21.3 ± 1.1</td>
</tr>
<tr>
<td>Mono-(2-ethyl-5-carboxypentyl) phthalate</td>
<td>35.2 ± 1.7</td>
</tr>
<tr>
<td>Mono-(2-ethyl-5-hydroxyhexyl) phthalate</td>
<td>24.8 ± 0.4</td>
</tr>
<tr>
<td>Mono-(2-ethyl-5-oxohexyl) phthalate</td>
<td>14.9 ± 0.4</td>
</tr>
<tr>
<td>Mono-(2-ethylhexyl) phthalate</td>
<td>4.13 ± 0.15</td>
</tr>
<tr>
<td>Mono-(3-carboxypropyl) phthalate</td>
<td>2.99 ± 0.20</td>
</tr>
<tr>
<td>Monobenzyl phthalate</td>
<td>8.37 ± 0.18</td>
</tr>
<tr>
<td>Monoethyl phthalate</td>
<td>94.5 ± 3.0</td>
</tr>
<tr>
<td>Mono-isobutyl phthalate</td>
<td>6.40 ± 0.28</td>
</tr>
<tr>
<td>Mono--n-butyl phthalate</td>
<td>10.6 ± 0.5</td>
</tr>
</tbody>
</table>
State Biomonitoring Cooperative Agreements

- **Technical support (2009+)**
  - Training
  - Site visits
  - Advisory services

- **Quality assurance programs (2012+)**
  - In-kind performance testing
    - Phthalates & other plasticizers
    - PAHs
    - Environmental Phenols & PCPs
    - Pesticides
      - Universal Pesticides
      - Dialkyl Phosphates
    - PFCs
Exposure to Phthalates in the United States

- Most Americans (6+ years) are exposed

<table>
<thead>
<tr>
<th>Compound</th>
<th>Detection frequency (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>DEHP</td>
<td>100</td>
</tr>
<tr>
<td>DiNP</td>
<td>100</td>
</tr>
<tr>
<td>DiDP</td>
<td>99</td>
</tr>
<tr>
<td>BBzP</td>
<td>98</td>
</tr>
<tr>
<td>DnBP</td>
<td>94</td>
</tr>
<tr>
<td>DiBP</td>
<td>99</td>
</tr>
<tr>
<td>DEP</td>
<td>100</td>
</tr>
</tbody>
</table>

n=2,489
Is Americans’ Exposure to Phthalates Changing?

- Some exposures increased: DiBP (↑ 121%)
- Other exposures decreased: DBP (↓ -60%)

![Chart showing NHANES sampling cycle with urine metabolite levels (µg/L)]
Americans’ Exposures Change across Phthalates

- Some exposures increased: DiNP (↑265%)
- Other exposures decreased: DEHP (↓-67%)
- Legislative actions and public scrutiny

![Graph showing Urine metabolite levels (µg/L) across NHANES sampling cycle 2005-2012 for DiNP and DEHP.](https://www.cdc.gov/exposurerreport)
Other Plasticizers: DINCH

- Phthalate alternative introduced in Europe in 2002
- DEHP replacement
  - Toys, medical devices, food packaging
- Metabolites as exposure biomarkers

Adapted from Koch et al. Arch Toxicol 2013
Are Exposures to DINCH Changing?

- Convenience U.S. adult sampling (2000-2012)
  - DINCH metabolites
  - Undetected in 2000-1
  - Increasing detection frequency after 2001
  - Increasing concentrations

- Similar results observed in Germany

<table>
<thead>
<tr>
<th>Year</th>
<th>N</th>
<th>Detection frequency (%)</th>
<th>50th (µg/L)</th>
<th>95th (µg/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2000</td>
<td>114</td>
<td>0</td>
<td>&lt;LOD</td>
<td>&lt;LOD</td>
</tr>
<tr>
<td>2001</td>
<td>57</td>
<td>0</td>
<td>&lt;LOD</td>
<td>&lt;LOD</td>
</tr>
<tr>
<td>2007</td>
<td>23</td>
<td>4</td>
<td>&lt;LOD</td>
<td>&lt;LOD</td>
</tr>
<tr>
<td>2009</td>
<td>118</td>
<td>8</td>
<td>&lt;LOD</td>
<td>0.5</td>
</tr>
<tr>
<td>2011</td>
<td>94</td>
<td>13</td>
<td>&lt;LOD</td>
<td>1.5</td>
</tr>
<tr>
<td>2012</td>
<td>121</td>
<td>19</td>
<td>&lt;LOD</td>
<td>1.4</td>
</tr>
</tbody>
</table>

LOD: 0.4 µg/L
### DINCH General Population Data

#### Germany
- **ESB**
  - 24-h urine samples
  - College students
  - 60 samples/year
  - 4 metabolites

#### USA
- **NHANES 2011-2**
  - Spot sample
  - 6+ years old
  - One metabolite (OH-MINCH)
  - Detection frequency: 24% (605/2489)
  - Range: <LOD (0.4 µg/L) to 168 µg/L
Are DINCH and Other Phthalates Replacing DEHP?

- DINP/DEHP trends in convenience samples
  - US & German general population
- DINCH & DINP may be replacing DEHP
  - Isomeric compounds
  - Starting with NHANES 2013-4, CDC will include another DINCH isomer
Monitoring Changes in Phthalates Exposures

- **Constantly evolving**
- **Identification of biomarkers**
  - In-vitro metabolism
  - In-vivo animal studies
  - Human studies
    - University of Bochum, Germany
- **Biomarkers choice**
- **Access to archived urine**
  - Convenience samples
  - General population samples
Many analytes can be measured simultaneously, but additional information is needed to demonstrate the utility of these analytes as exposure biomarkers.
### Selection of Exposure Biomarkers: DINP Example

- **DINP metabolites:** MNP (~2%) and MCOP (~11%)
- **MNP** (minor metabolite): insensitive biomarker of DINP background exposures

<table>
<thead>
<tr>
<th>MNP urinary concentrations</th>
<th>MCOP urinary concentrations</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Detectable</td>
</tr>
<tr>
<td>Detectable</td>
<td>347 (12.9%)</td>
</tr>
<tr>
<td>Non-detectable</td>
<td>2100 (82.4%)</td>
</tr>
<tr>
<td>Total</td>
<td>2447 (95%)</td>
</tr>
</tbody>
</table>

- Select most abundant/relevant biomarker to minimize exposure misclassification
  - 82.4% of persons classified as exposed to DINP are misclassified based on urinary concentrations of MNP only
Dibutyl Phthalates in NHANES

- NHANES 1999-2000: MBP (MnBP + MiBP)
- NHANES 2001-12: MnBP & MiBP
- NHANES 2013+: MnBP, MiBP, 3OH-MnBP, 2OH-MiBP

Adapted from Koch et al. Arch Toxicol 2012
Nitrocellulose plasticizer
- Plastic film, inks & wood coatings

Testicular toxicant in rats

In-vivo metabolism in rats
- Nine rats
- Single oral dose (500 mg/Kg)
- Urine collected 24-h & 48-h after dose

**DPP metabolite median urinary concentrations (µg/mL)**

<table>
<thead>
<tr>
<th>Metabolite</th>
<th>24-h post dose</th>
<th>48-h post dose</th>
</tr>
</thead>
<tbody>
<tr>
<td>MPP</td>
<td>222</td>
<td>75</td>
</tr>
<tr>
<td>MHPP</td>
<td>993</td>
<td>191</td>
</tr>
<tr>
<td>MCBP</td>
<td>168</td>
<td>29</td>
</tr>
</tbody>
</table>
Human Exposure to Dipentyl Phthalate (DPP)

- Forty-five spot samples
- Anonymously collected in 2009 from adults
  - Relatively low detection frequency for MHPP (specific)
  - No correlation between MHPP & MCBP/MCPP
- Limited exposure to DPP in US adults

### DPP Metabolite Urinary Concentrations

<table>
<thead>
<tr>
<th>Metabolite</th>
<th>% Detection</th>
<th>Min (µg/L)</th>
<th>Max (µg/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>MPP</td>
<td>0</td>
<td>NA</td>
<td>NA</td>
</tr>
<tr>
<td>MHPP</td>
<td>29</td>
<td>&lt;LOD</td>
<td>8</td>
</tr>
<tr>
<td>MCBP</td>
<td>4</td>
<td>&lt;LOD</td>
<td>221</td>
</tr>
<tr>
<td>MCPP</td>
<td>13</td>
<td>&lt;LOD</td>
<td>40</td>
</tr>
</tbody>
</table>
What Exposure Biomarkers Should We Measure?

- **Analytical method**
  - Can we add more analytes?
  - Instrumentation
    - DiDP vs Bis-(2-propylheptyl)phthalate (DPHP)

- **Toxicokinetics**
  - Abundance
  - Specificity

- **Target population**
  - Exposures can be population-specific
    - Age-dependent

- **Nature of exposure**
  - Background vs specific exposures
Americans are exposed to phthalates

Market changes in commercial formulations
  - Introduction of replacement chemicals
    - Phthalates (e.g., DiNP)
    - Non-phthalates (e.g., DINCH)
  - Changing exposures

Biomonitoring & biomarkers toxicokinetics
  - Specificity
  - Abundance

Method adequate for intended purpose

Banking of urine
  - Trends evaluation
Future Work

- Continue NHANES & studies on targeted populations
  - Track exposures to “legacy” & replacement chemicals
  - Fill in data gaps to better understand temporal trends and underlying reasons

- Identify & incorporate phthalate and phthalate replacement biomarkers
Acknowledgements

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Our collaborators
THANK YOU!

For more information please contact Centers for Disease Control and Prevention

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The findings and conclusions in this report are those of the authors and do not necessarily represent the official position of the Centers for Disease Control and Prevention.