Novel Approaches for Chemical Mixture Risk Assessment

Carl-Gustaf Bornehag, PhD
Professor in Public Health Sciences

Karlstad University, Karlstad, Sweden
Icahn School of Medicine at Mount Sinai, New York City, USA

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## Risk assessment strategies

<table>
<thead>
<tr>
<th>Evidence of risk</th>
<th>Single compound approach</th>
<th>Hazard Quotient (HQ)</th>
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<tr>
<td>Exposures</td>
<td>Single chemicals of interest</td>
<td>Exposure/RfDs &gt; 1</td>
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<tr>
<td>Epidemiological</td>
<td>Qualitative evaluation</td>
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<td>Experimental dose response (BMD, PODs, RfDs, etc.)</td>
<td>Guideline values (PODs/RfDs for humans; BE/HBM values)</td>
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**Risk assessment strategies**
## Published guideline values for urinary DBP, BBzP, DEHP and DINP

<table>
<thead>
<tr>
<th>Diester</th>
<th>Metabolites in Mixture S0</th>
<th>Observed concentration in SELMA (N=2,313)</th>
<th>Published HBM or BE Values</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>95&lt;sup&gt;th&lt;/sup&gt; percentile</td>
<td>99&lt;sup&gt;th&lt;/sup&gt; percentile</td>
</tr>
<tr>
<td>DBP</td>
<td>MBP</td>
<td>239</td>
<td>590</td>
</tr>
<tr>
<td>BBzP</td>
<td>MBzP</td>
<td>102</td>
<td>257</td>
</tr>
<tr>
<td>DEHP</td>
<td>Sum of 4 metabolites</td>
<td>11</td>
<td>35</td>
</tr>
<tr>
<td>DINP</td>
<td>Mono-carboxyoctyl phthalate (MCOP)</td>
<td>77</td>
<td>261</td>
</tr>
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<td>Human relevance Co-exposures</td>
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<td><strong>Evidence of risk</strong></td>
<td>Epidemiological Qualitative evaluation</td>
<td>Qualitative evaluation</td>
<td>Selection of bad actors (WQS, etc.)</td>
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<td>Experimental dose response (BMD, PODs, RfDs, etc.) Guideline values (PODs/RfDs for humans; BE/HBM values)</td>
<td>Guideline values (PODs/RfDs for humans)</td>
<td>BMD for reference mixture(s)</td>
</tr>
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<td><strong>Risk evaluation</strong></td>
<td>Hazard Quotient (HQ) Exposure/RfDs &gt; 1</td>
<td>Hazard Index (HI) ΣHQ &gt; 1</td>
<td>Test for sufficient similarity SMRI SMRI &gt; 1</td>
</tr>
</tbody>
</table>
REAL LIFE CHEMICAL MIXTURES BASED ON URINE/SERUM LEVELS FROM +2,300 PREGNANT WOMEN IN THE SELMA STUDY
Whole mixture approach
four steps for risk assessment of chemicals
integrating human epidemiology and experimental toxicology

1. Identification of bad actors (mixtures) for health effects in epidemiological data
2. Composition of reference mixtures from population data for experimental evaluations
3. Experimental tests (in cells and animals) of reference mixtures for dose-response
4a. Test for sufficient similarity with the reference mixture (%)  
4b. For sufficient similar subgroups; test for extreme mixing proportions, SMRI>1 (%)  
4c. Demonstrate if health effects are associated with SMRI (adj risk, 95% CI)

Sufficient Similarity Approach (SMACH)

Mixture S  
Mixture N  
Mixture G
Natural hormones

Estrogen
Testosterone
Thyroids, etc.

Health and development

20/54 EDCs
(N=+2,300)

Mixtures

Metabolism and growth (G)
Sexual development (S)
Neurodevelopment (N)
Analysis strategy in three steps for Mixture 0

1. Identification of **bad actors** for the three health domains
   - Weighted quantile sum (WQS) regression (Carrico, 2015)
Natural hormones

- Estrogen
- Testosterone
- Thyroids, etc.

20/54 EDCs
(N=+2,300)

Mixture G
Macrosomia
IUGR***
Median
IUGR

Body weight

Birth  Wean  Adult

The fetal period is important for chronic diseases later on in life, e.g., hypertonia, diabetes, cardiovascular diseases...

David Barker (1938-2013)
Analysis strategy in three steps for Mixture 0

1. Identification of bad actors for the three health domains
   – Weighted quantile sum (WQS) regression (Carrico, 2015)

2. Estimation of serum levels of bad actors
   – Estimation of daily intake (DI) of urinary based bad actors (Koch et al., 2007)
   – Using toxicokinetic models (Fromme et al., 2007) estimating the plasma concentrations from DI
   – The PFASs were measured directly in serume levels
Analysis strategy in three steps for Mixture 0

1. Identification of **bad actors** for the three health domains
   - Weighted quantile sum (WQS) regression (Carrico, 2015)

2. Estimation of **serum levels of bad actors**
   - Estimation of daily intake (DI) of urinary based bad actors (Koch et al., 2007)
   - Using toxicokinetic models (Fromme et al., 2007) estimating the plasma concentrations from DI
   - The PFASs were measured directly in serum levels

3. Establishment of **relevant mixtures**, to be evaluated in experimental studies in cell and animal models
   - Estimation of mixing proportions of bad actors using serum levels in +2,300 pregnant women in SELMA
   - The mixing proportions were calculated in molar units across the chemicals
Natural hormones
Estrogen
Testosterone
Thyroids, etc.

Sexual development & fertility

15 COMPOUNDS
Phthalates
Phenols
PFASs
# Mixture S0

<table>
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<tr>
<th>Identification of bad actors among 20 EDCs</th>
<th>Determination of a typical mixture of bad actors</th>
<th>Composition of a reference mixture</th>
</tr>
</thead>
<tbody>
<tr>
<td>Using Weighted Quantile Sum (WQS) regression</td>
<td>Using geometric mean serum levels (mol/L) in +2,300 SELMA mothers</td>
<td>Dosing 0.1X, 1X, 10X, 100X where 1X refers to SELMA</td>
</tr>
<tr>
<td><strong>DBP</strong></td>
<td>2.3 E-08</td>
<td>33%</td>
</tr>
<tr>
<td><strong>BBzP</strong></td>
<td>1.1 E-08</td>
<td>16%</td>
</tr>
<tr>
<td><strong>DEHP</strong></td>
<td>1.5 E-08</td>
<td>21%</td>
</tr>
<tr>
<td><strong>DiNP</strong></td>
<td>2.1 E-08</td>
<td>30%</td>
</tr>
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</table>
Test for dose-response relationship between Mixture S and AGD/BW in male mice

With a benchmark response (BMR) of a 5% decline in AGD/BW, the estimated benchmark dose (BMD) was 0.49 on the log scale

$10^{0.49} - 1 = 2.1X$ of "typical" SELMA exposure

National and Kapodistrian University of Athens, Athens, Greece
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Mixture S
Mixture N
Mixture G
4-5% (1.6-1.8 mm) reduction of AGDAs in baby boys in SELMA

Bornehag et al., 2015
83% of the SELMA women (N=1,916 out of 2,313) had sufficiently similar mixing proportions to Mixture S.
4b. Similar Mixture Risk Index (SMRI)

For the set of sufficiently similar mixtures, roughly 7% of the SELMA women have concentrations extreme relative to the BMD (SMRI>1), corresponding to about 6% of the total population of 2,313 pregnant women.
4c. Association between SMRI and AGD in baby boys

Adjusted* AGD was 5.9 mm shorter in the 4th vs. 1st quartile of SMRI (p=0.045)

* Adjusted for gestational age at exposure, weight at evaluation, and creatinine
Conclusions

With a whole mixture approach, we could find a higher rate of pregnant women under risk (13%) when comparing with more traditionally models of additivity (HI) (3%), or a compound-by-compound strategy (1.6%), which is the most used risk assessment procedure.

Bornehag, Kitraki, Panagiotidou, Stamatakis, Ruden, Shu, Lindh, Ruegg, Gennings

A novel approach to chemical mixture risk assessment - Linking data from population based epidemiology and experimental animal tests by the use of new statistical tools

Risk Analysis, in revision
New approaches for risk assessment of chemical mixtures

Human pregnancy cohort data (exposures and health effects)

Mixture Desirability Function (MDF)

- Non-linear statistical models evaluating regulatory guideline values
  - Acceptable concentration ranges (ACR)

A whole mixture approach

- Mixtures of bad actors tested in experimental settings
  - Similar mixture approach (SMACH) SMRI>1 (%)
Incorporating regulatory guideline values in analysis of epidemiology data

Chris Gennings\textsuperscript{a} \textsuperscript{,} Huan Shu\textsuperscript{b}, Christina Rudén\textsuperscript{b}, Mattias Öberg\textsuperscript{c}, Christian Lindh\textsuperscript{d}, Hannu Kiviranta\textsuperscript{e}, Carl-Gustaf Bornehag\textsuperscript{a, f}


draft

Highligths

We introduce a new class of models that include the regulatory concept of “acceptable concentration range” (ACR)

These ACR models complement current risk assessment methods by estimating guideline values using human biomonitoring data

The results suggest that chemical-by-chemical approaches underestimate risk by a factor that range from 1 to 100 for different chemicals
On-going work

Analyses of all experimental data for Mixture 0

Analysis of Mixture 1 data
54 chemicals
Health outcomes at 7 years of age

Assess generalizability
Test for sufficient similarity in existing biomonitoring data (HB4EU)
Development of mixture assessment factors (MAFs) using ACR
Thank you!