NIEHS mixture workshop 26-27 Sept 2011

Mixture effects and epidemiology: The need for unbiased approaches in exposure assessment

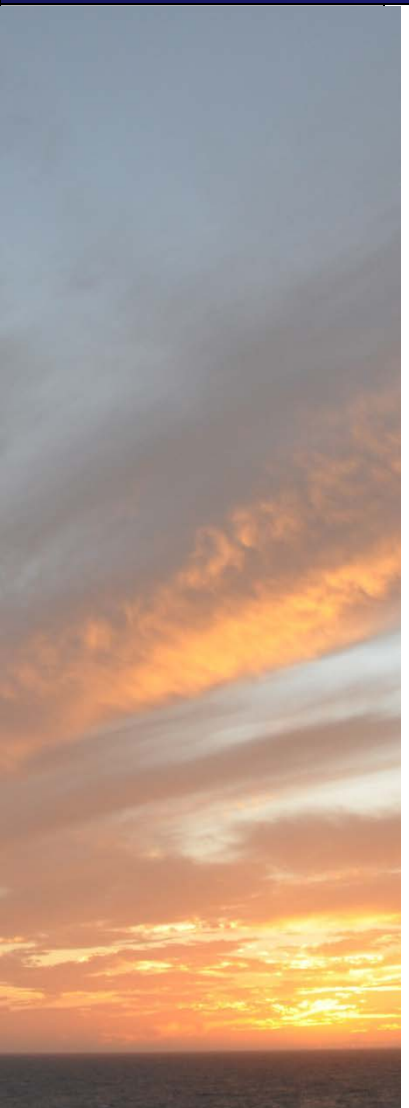
Andreas Kortenkamp

Institute for the Environment, Brunel University, London

Evidence from mixture toxicology

- **Predictability**: Mixture effects predictable when toxicity of components known
- **Dose range**: Mixture effects at low (effect) doses
- **Tools**: Dose addition a good approximation

The subversive influence of mixture toxicology



- ADI (TDI) – sufficiently protective?
- Exposures – are they defined?
- Hazards – tested at all?

When is a mixture “safe”?

The case of dose addition

$$\frac{\text{Intake}_1}{\text{Tolerable Daily Intake}_1} + \frac{\text{Intake}_2}{\text{Tolerable Daily Intake}_2} < 1$$

Mixture effect equal to effect at TDI if every component is present at **TDI / n**

How many mixture components are we dealing with?

When is a mixture "safe"?

The case of independent action

Independent action

$$E_{1,2,..n} = 1 - [(1-e_1)(1-e_2)...(1-e_n)]$$

100 agents with zero effect:	joint effect = 0
100 agents with 1% effect:	joint effect = 63%
100 agents with 0.1% effect:	joint effect = 9.5%

Is an ADI always a zero effect?

Cumulative risk assessment: restrictions

- **Effect criterion**: *in vivo* evidence of anti-androgenicity
- **Exposure information** available

Known knowns (16 chemicals)

Chemical	TDI	Exposure		HQ	HQ	HI %	HI %
		Median	High	median	high	median	high
DBP	0.1	0.002	0.006	0.0200	0.0600	12.6	7.3
DiBP	0.2	0.0015	0.001	0.0075	0.0050	4.7	0.6
BBP	0.33	0.0005	0.004	0.0015	0.0121	1.0	1.5
DEHP	0.05	0.0027	0.0036	0.0540	0.0720	34.0	8.8
DPP	0.05	0	0	0.0000	0.0000	0.0	0.0
DiNP	1.5	0.0006	0.005	0.0004	0.0033	0.3	0.4

Known knowns (16 chemicals)

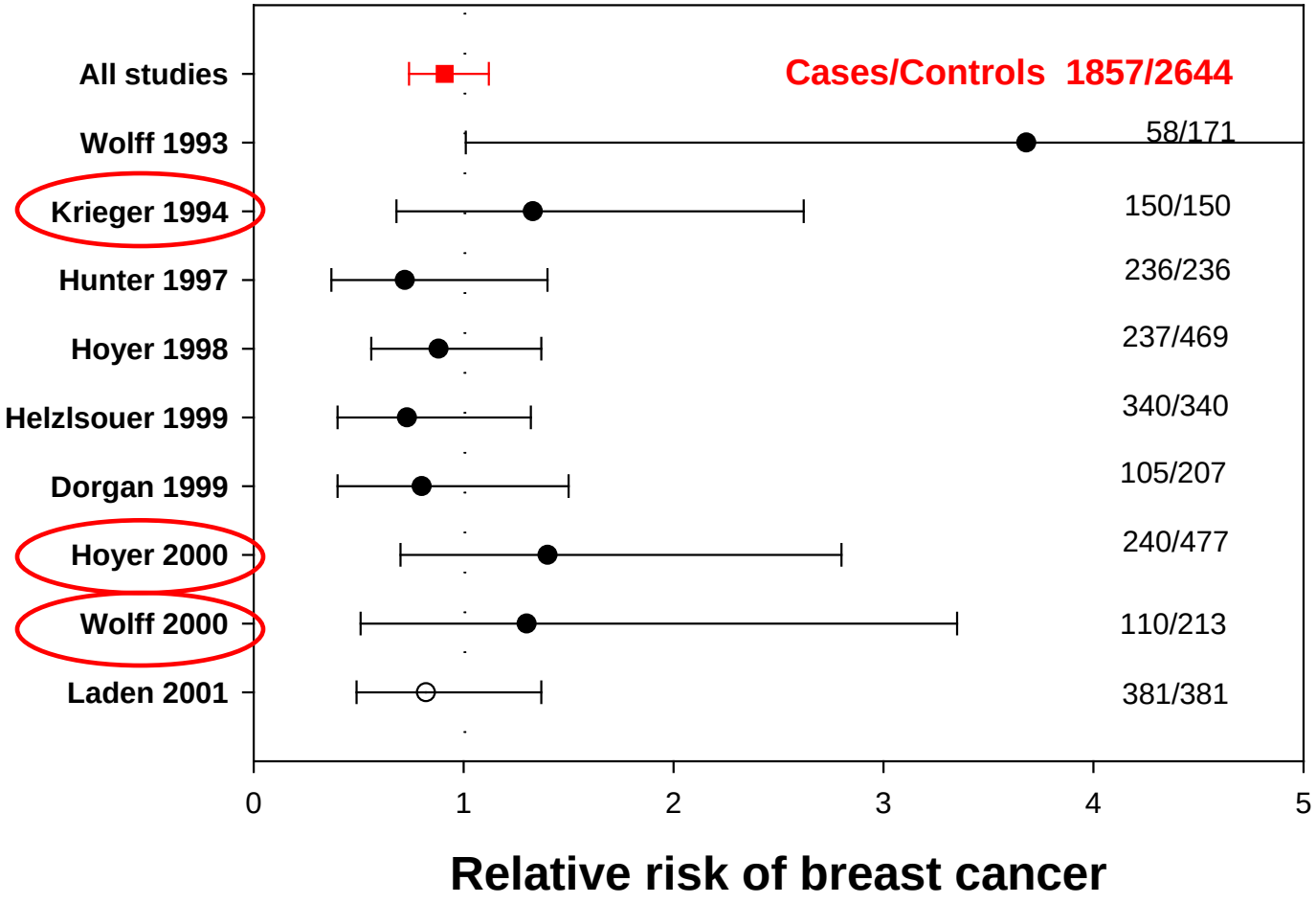
- 7 chemicals explain > 90% of expected combination effect
- But what will the results be with knowledge of “unknowns”?

Kortenkamp and Faust (2010)
Internat J Androl **33**, 463

Organochlorines and breast cancer – a meta-analysis

Outcome of prospective studies of *p,p'*-DDE and pooled analysis

Lopez-Cervantes *et al.* (2004), EHP **112**, 207



Environmental epidemiology results

On organochlorines and breast cancer

"…these results should be regarded as strong evidence to discard the putative relationship between *p,p'*-DDE and breast cancer risk."

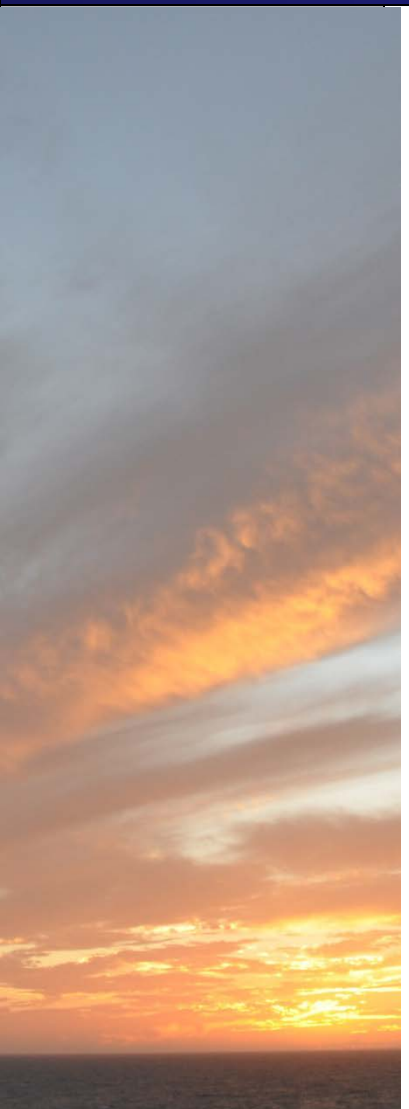
Lopez-Cervantes *et al.* (2004) EHP **112**, 207

On organochlorines and hypospadias and cryptorchidism

"This study does not support an association of DDT or DDE with hypospadias or cryptorchidism."

Bhatia *et al.* (2005) EHP **113**, 220

Unbiased exposure assessment strategies – the exposome



Steve Rappaport (2011) Tox Lett (in press)

However, with few exceptions, the identities of major environmental toxicants and their roles in causing chronic diseases have not been addressed.

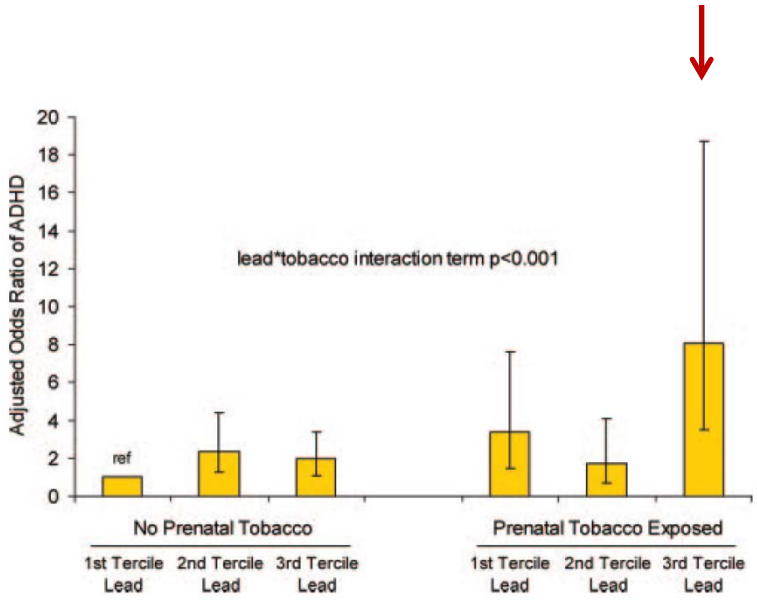
Given the poor state of knowledge about health-impairing environmental exposures, epidemiologists pursue narrow hypotheses that largely skirt disease etiology in favor of known environmental risk factors, even when the attributable risks are small. Although such hypothesis-driven studies confirm some environmental sources of disease, they offer only fragments to our understanding of the major causes and mechanisms of chronic diseases.

Possible goals of mixture epidemiology

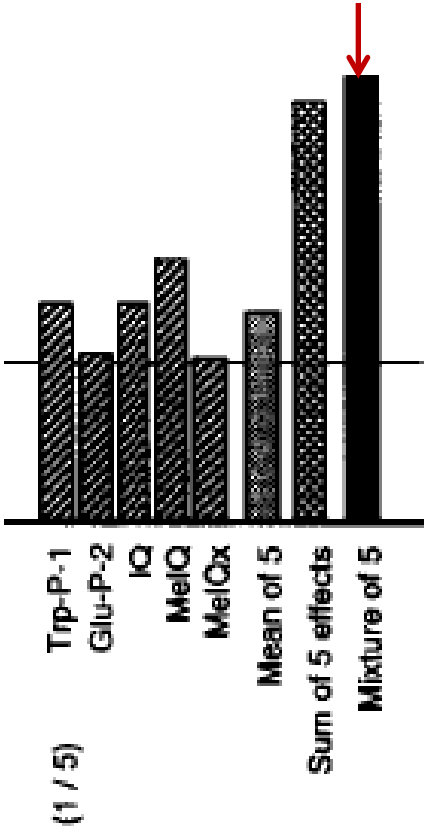
Cohort studies	• Health impact of known combinations of chemicals (chemicals –> effects)
Case-control studies	• Impact of chemical exposures on diseases (disease –> chemicals)

Mixtures and epidemiology

"Synergisms" in mixtures

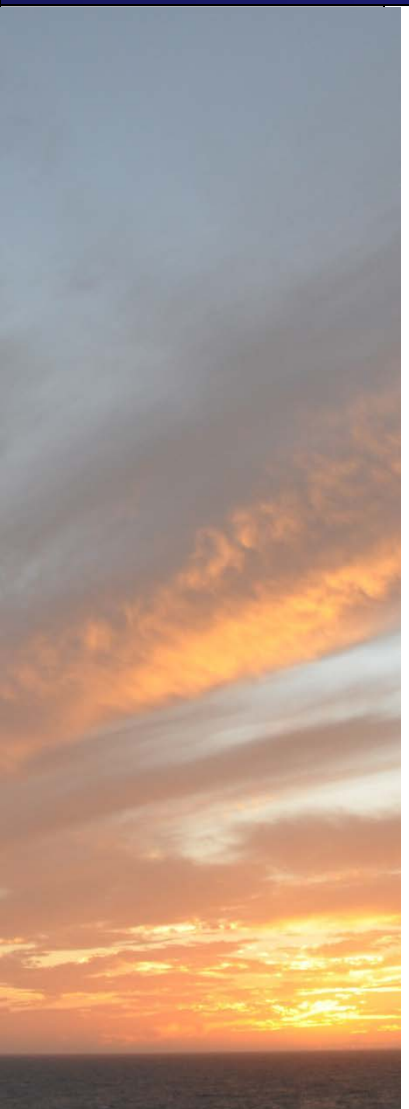


Froehlich (2009)



Hasegawa (1996)

Mixtures and epidemiology?

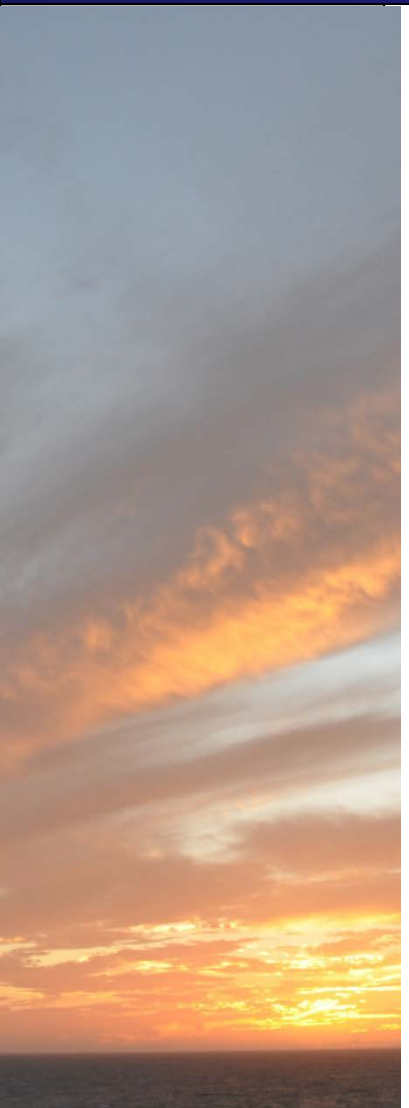


- Impact of chemical exposures on diseases (disease –> chemicals)
- Measuring multiple chemicals?
- Which chemicals (what is the hypothesis?

$$M_1 + M_2 + M_3 + M_4$$

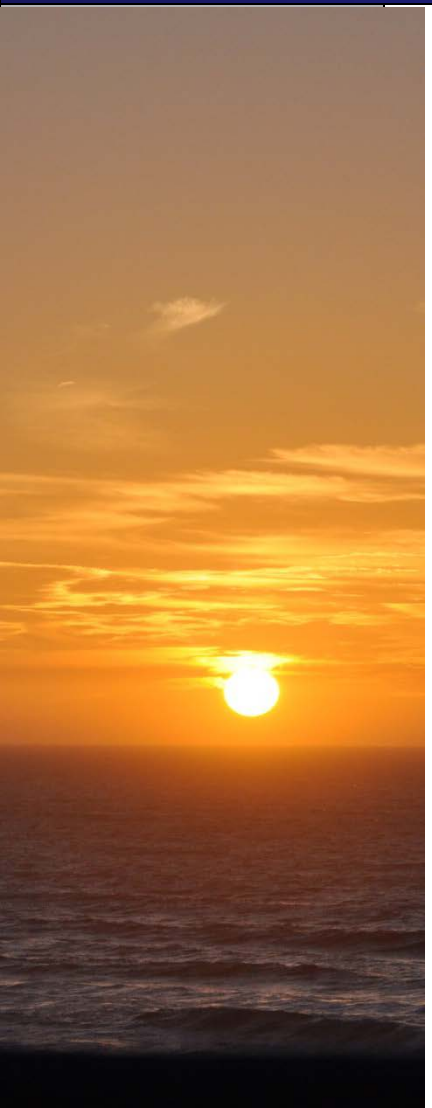
$$W_1 M_1 + W_1 M_2 + W_1 M_3 + W_1 M_4$$

Mixtures and epidemiology?



- Aggregate **prevalence** and **potency**
- Where should potency estimates come from?

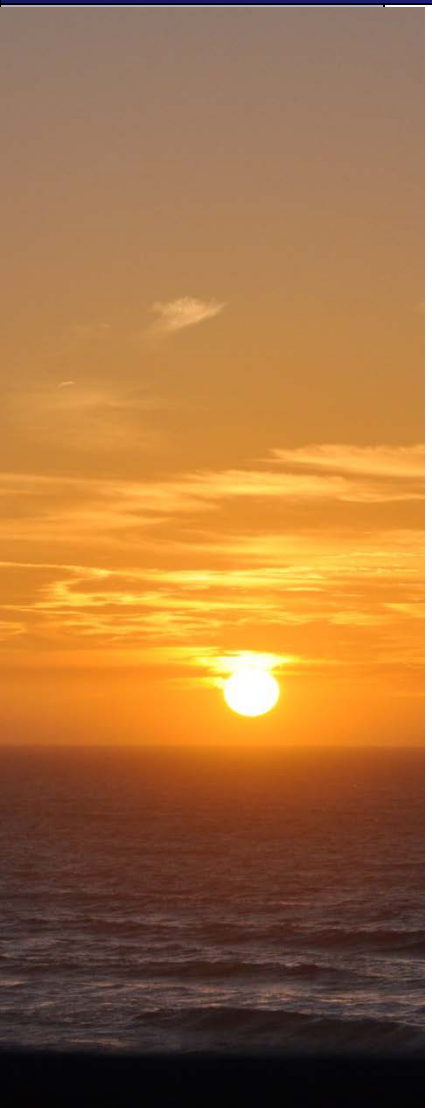
Hazard Quotients and Indices (approach 1)



$$HI = \sum_{i=1}^{n} \frac{EL_i}{AL_i}$$

- Exposure levels (EL)
- Acceptable levels for single chemicals (AL): NOAEL/UF
- Total number of chemicals (n)
- Correlate sum of hazard quotients with health outcomes

Data requirements (approach 1)



- **Exposure levels**: back-calculation from measured conc in tissues
- Toxicokinetic models
- **Potency**: from *in vivo* animal models
- **Example**: phthalate intake from urinary metabolite levels
- **Advantage**: compatibility with regulatory risk assessment
- **Disadvantage:** Data-hungry

Hazard Quotients and Indices (approach 2)

$$HI = \sum_{i=1}^{n} \frac{EL_i}{AL_i}$$

- EL: Tissue concentration (measured)
- AL: Effect conc in *in vitro* assay
- Total number of chemicals (n)

Data requirements (approach 2)

- Measured conc in tissues
- **Potency**: from suitable *in vitro* assay
- **Example**: *in vitro* AR antagonists, *in vitro* ER agonists
- **Advantage**: easier to apply
- **Issue**: *in vitro / in vivo* extrapolations – relevance debatable

Biomarkers of exposure

Ibarluzea *et al.* (2004) Cancer Causes Contr **15**, 591

16 individual organochlorines in adipose tissue: No differences

Measure of total estrogenicity (E-Screen) **of organochlorines in adipose tissue** (lean women)**:**

Estrogenicity	Odds ratio (CI)
0.25	1
0.26-41	1.12 (0.5-2.52)
41-197	1.58 (0.7-3.58)
>197	**2.44 (1.03-5.78)**

Acknowledgements

- Financial support from European Union - *contamed*
- UK Food Standards Agency

- www.nectarcluster.info

Thank you