New Technologies and Approaches in Toxicity Testing and Risk Assessment

Netherlands Society of Toxicology Meeting
June 19, 2014

Rusty Thomas, Ph.D.
Director, National Center for Computational Toxicology
(Thomas.Russell@epa.gov)

Current System is Antiquated and Inefficient

Acute toxicity studies (LD_{50}) developed to standardized batches of pharmaceuticals

Genotoxicity assays developed

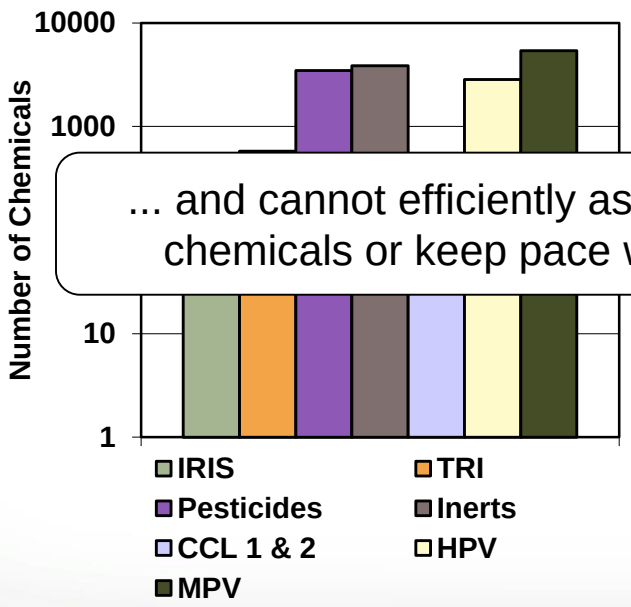
Continuous breeding studies for reproductive toxicity

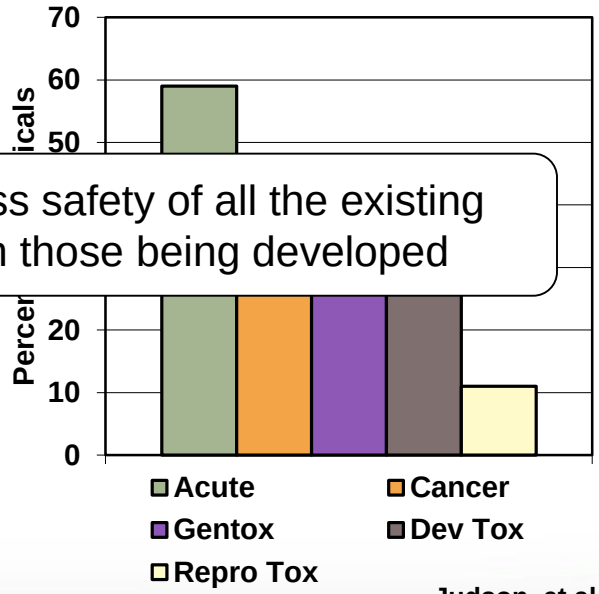
1920 | 2010

Current testing paradigm does not incorporate advances in technology and does not provide mechanistic data

Draize test introduced for eye irritants

Rodent cancer bioassay introduced

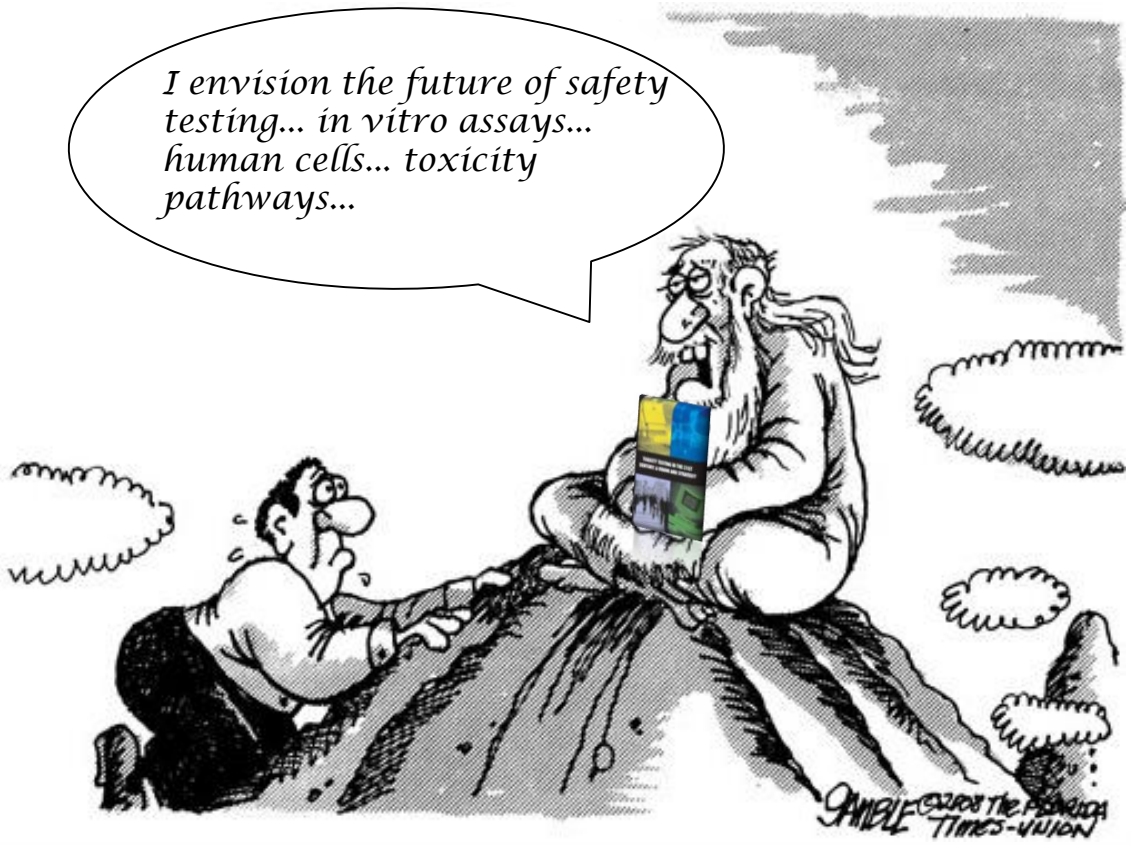




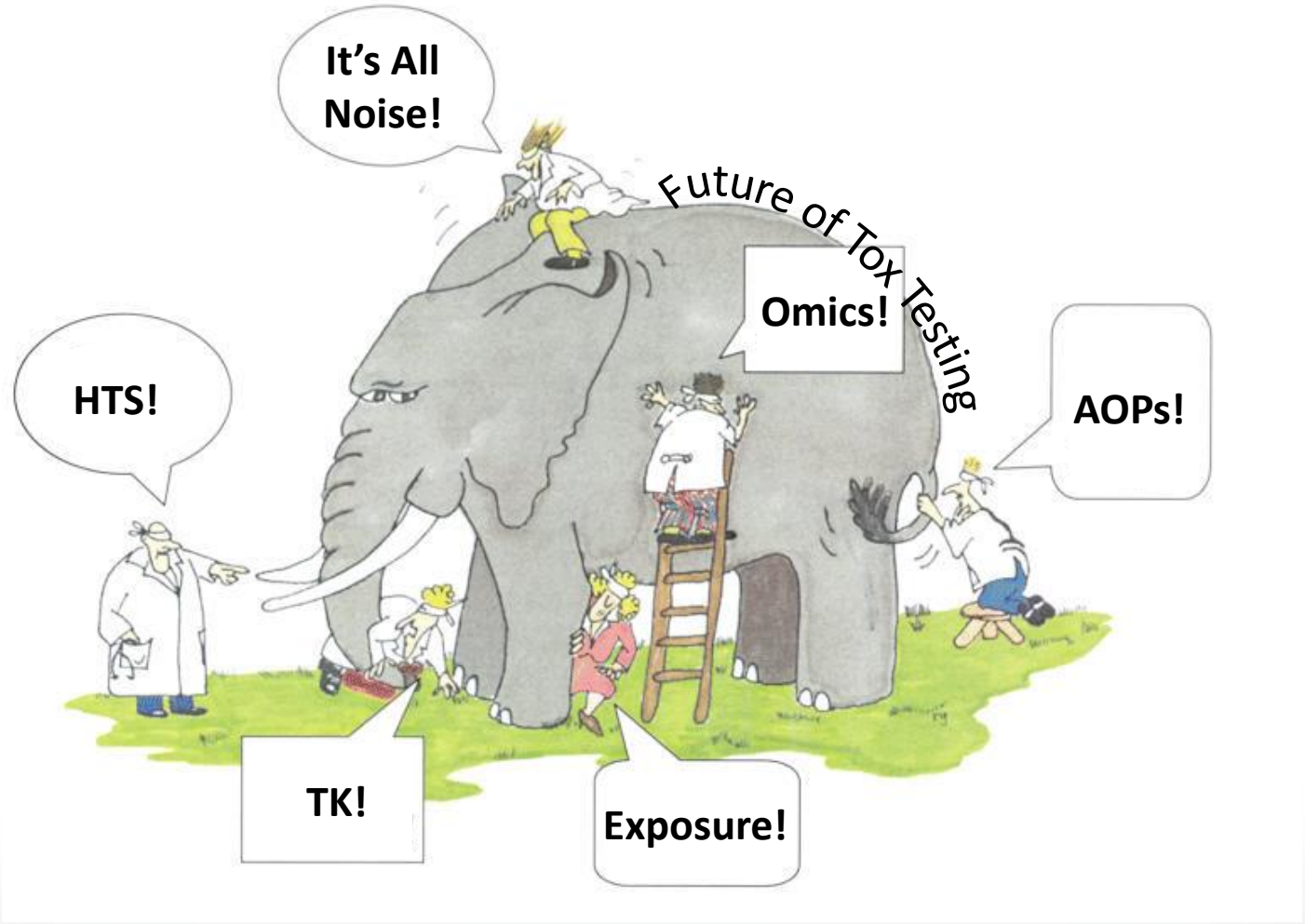
... and cannot efficiently assess safety of all the existing chemicals or keep pace with those being developed

Judson, et al *EHP* (2010)

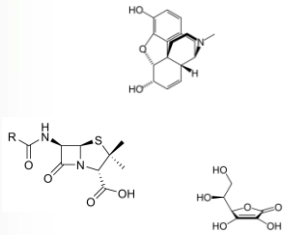
In 2007, NRC Transformed Toxicology with a Future View

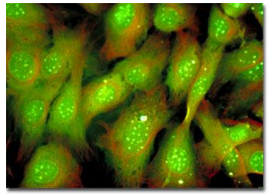


Let's Start from a Common Understanding



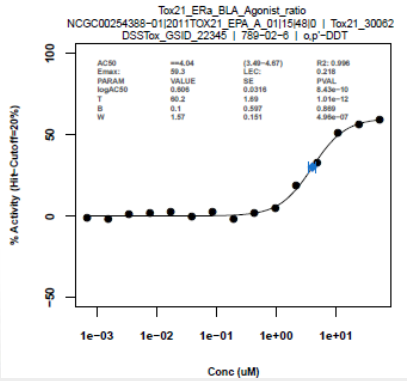
It is All About High-Throughput Screening (ToxCast)…





1,800 Chemicals in Concentration Response

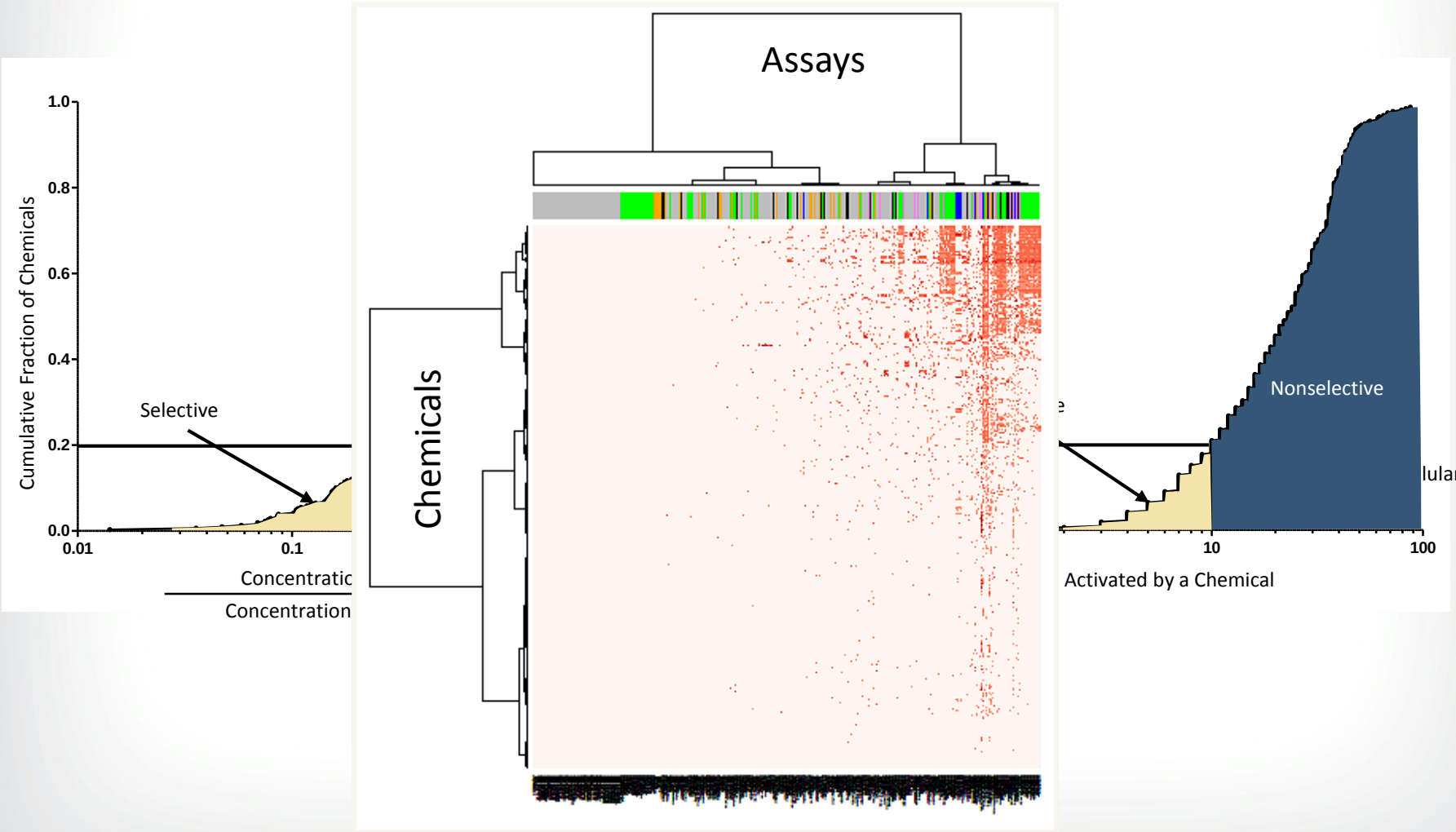
~300 Phase I
~700 Phase II
~800 E1K



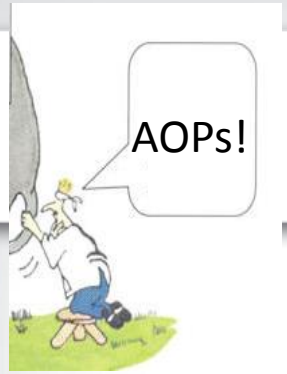
~700 Biochemical and Cell-based High-throughput Screening Assays

(327 genes and 293 pathways)

What Did High-Throughput Screening Tell Us?



Thomas *et al., Tox Sci.,* 2013



In Vitro Assay Selectivity as a Starting Point for Chemical MOAs/AOPs

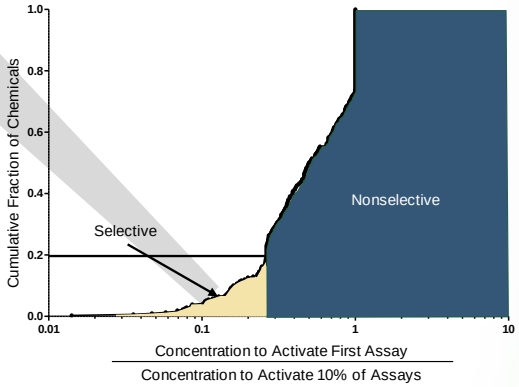
Selective Chemical

Selectively Activated *In Vitro* Assays

Define Mode-of-Action

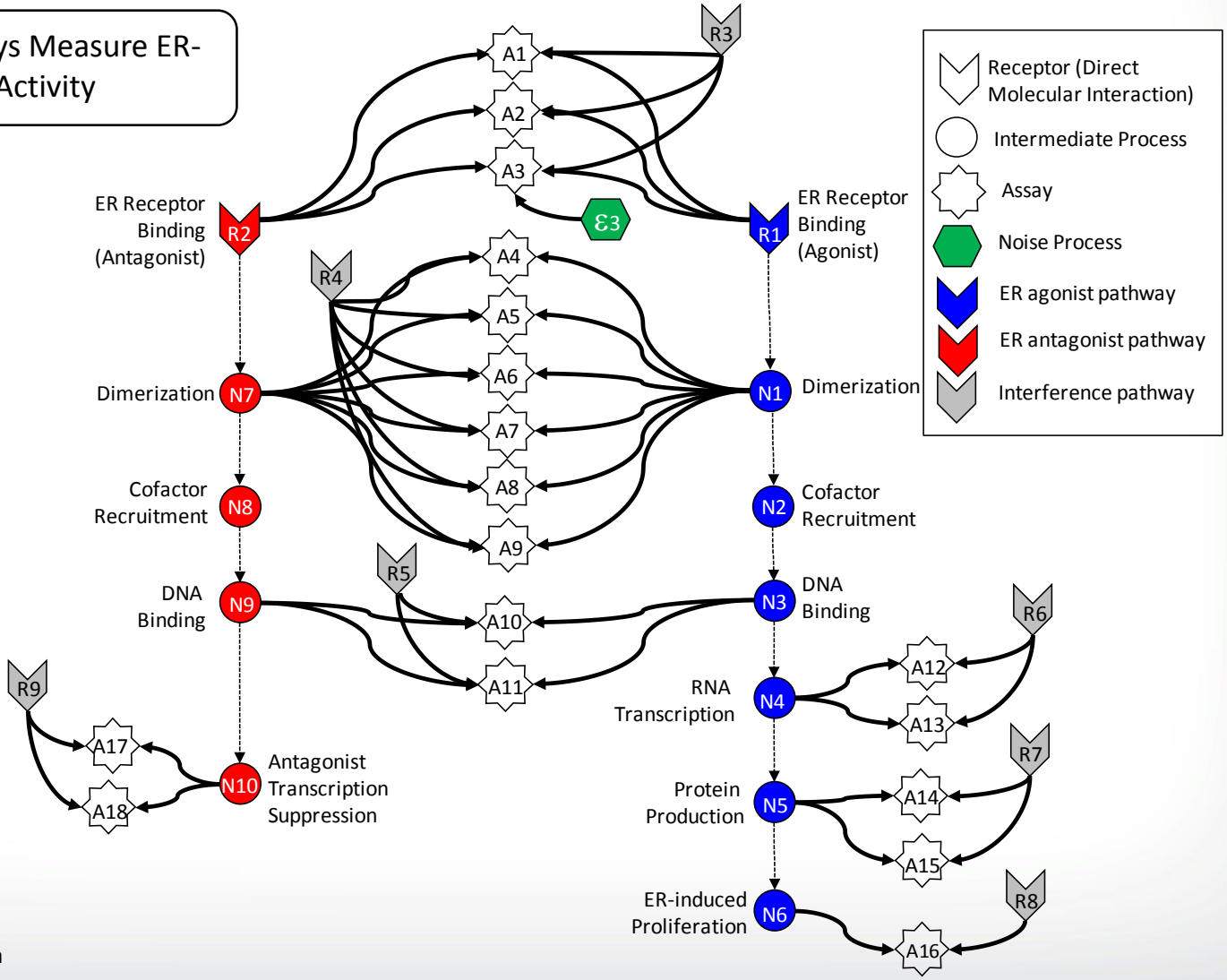
Confirm Human Relevance and Derive Point-of-Departure

Key Events



Combining Results from *In Vitro* Assays to Assess Estrogen-Related Activity

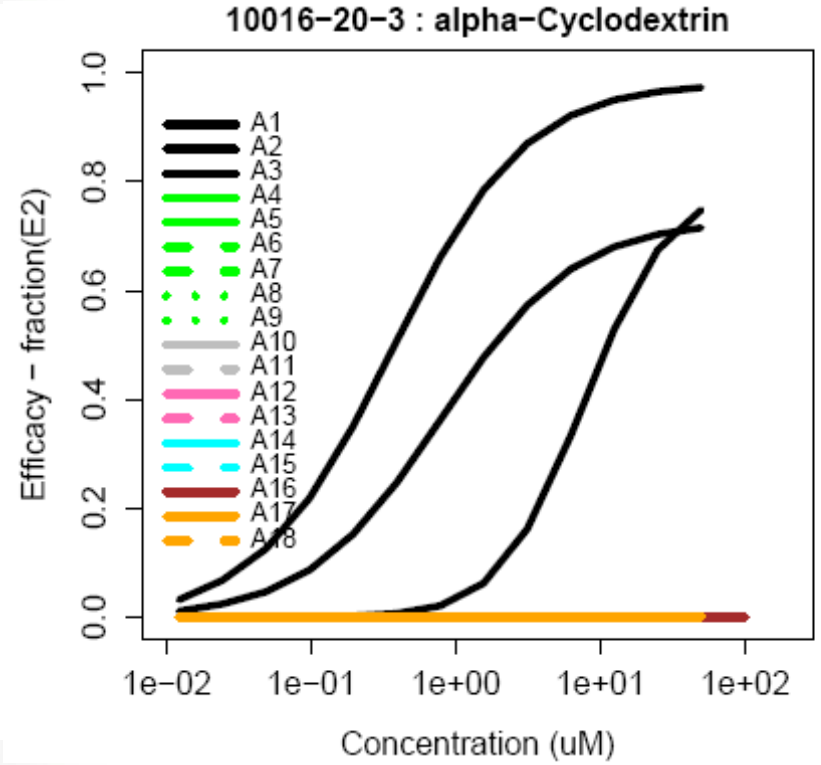
18 *In Vitro* Assays Measure ER-Related Activity



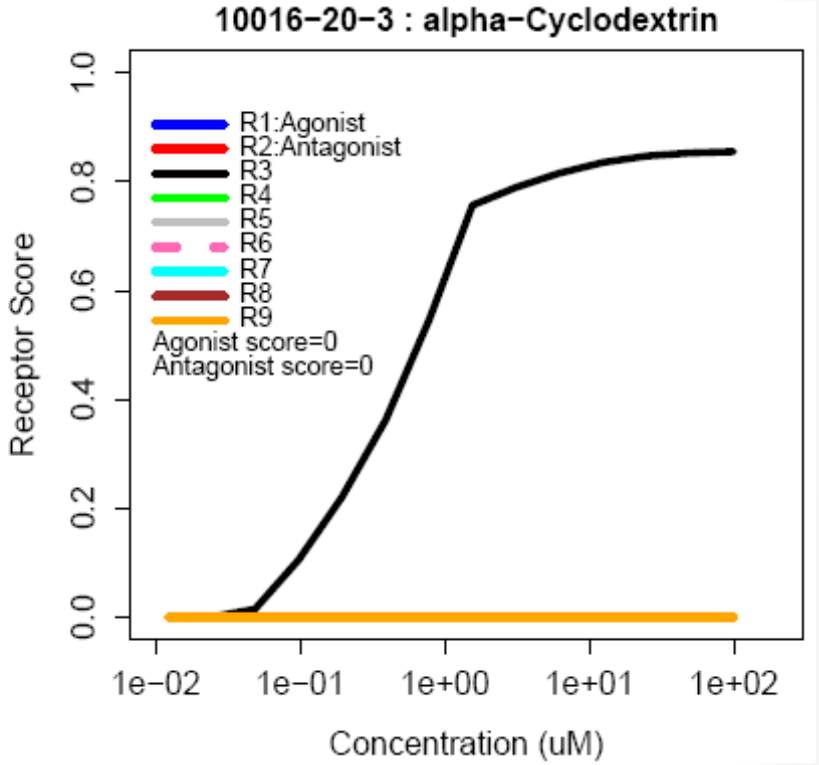
Judson *et al.*, Unpublished Data

Pathway-Based Modeling to Assess Specific Activity Versus Interference

Assays

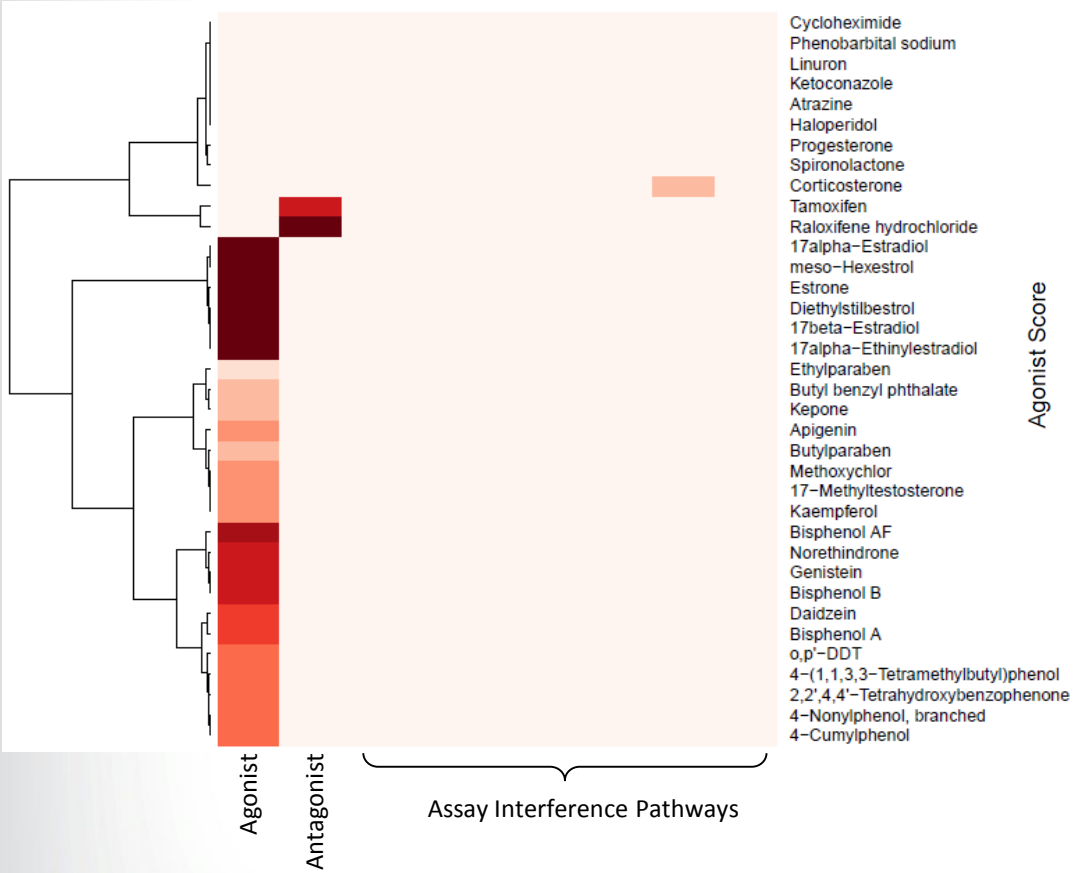


"Receptors"

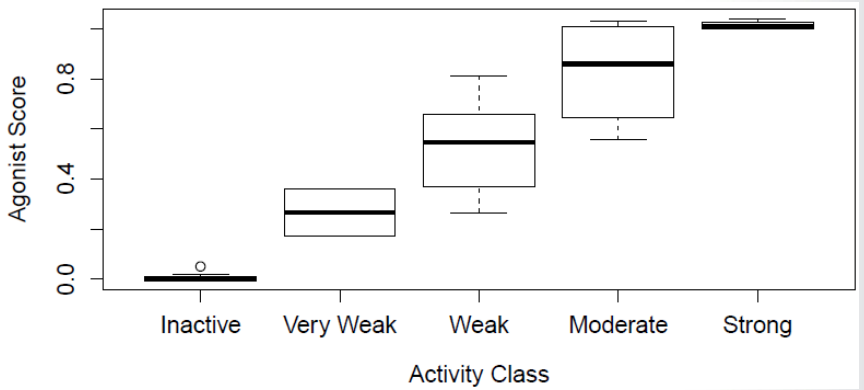


Judson *et al.*, Unpublished Data

Pathway-Based Modeling to Assess Specific Activity Versus Interference

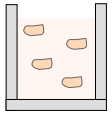


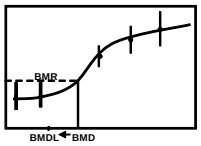
Heat Map of Model Scores for Reference Chemicals



Judson *et al.,* Unpublished Data

What Have We Learned About the Non-Selective Chemicals?

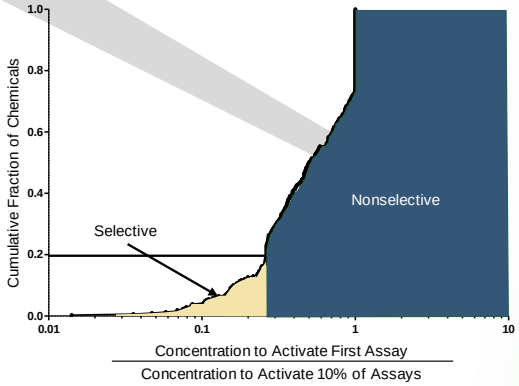




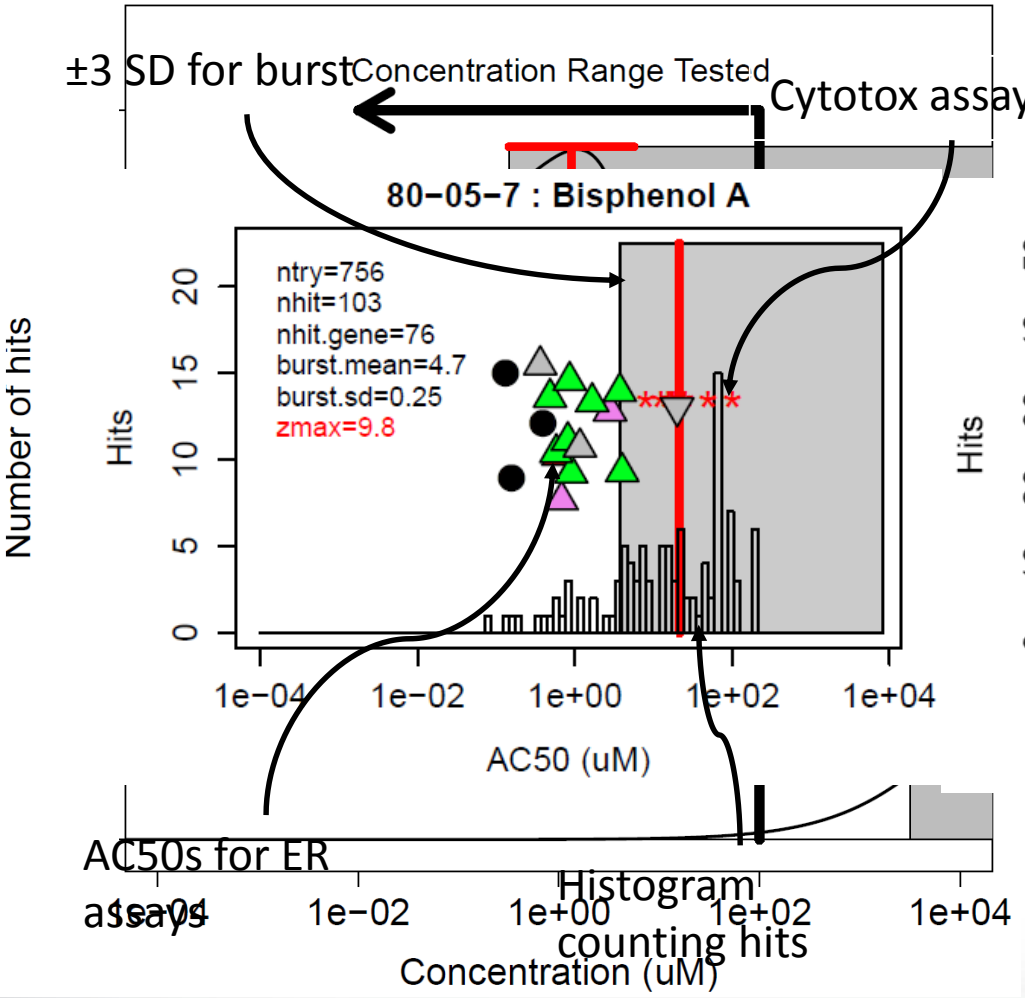
Nonselective Chemical

????????

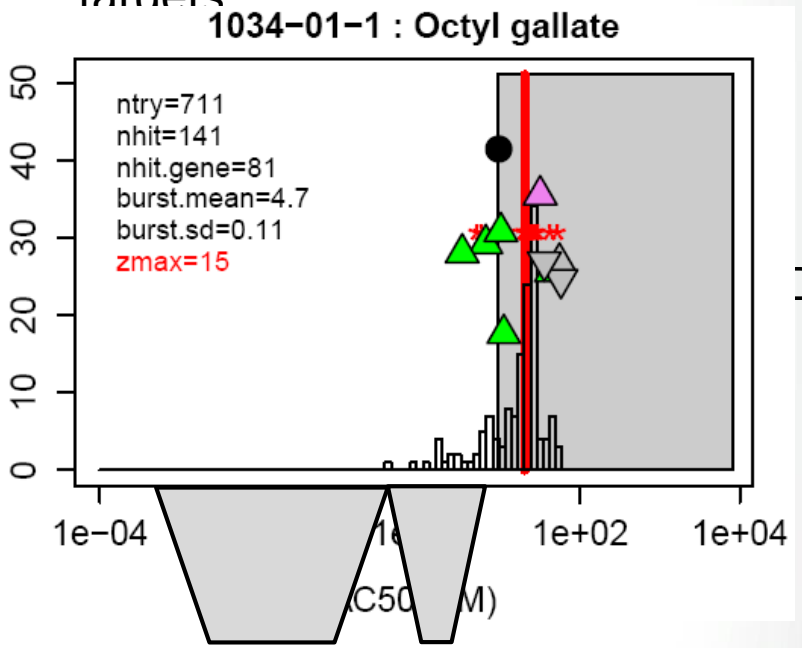
Define Point-of-Departure



Non-Selectivity Closely Aligned with Cytotoxicity



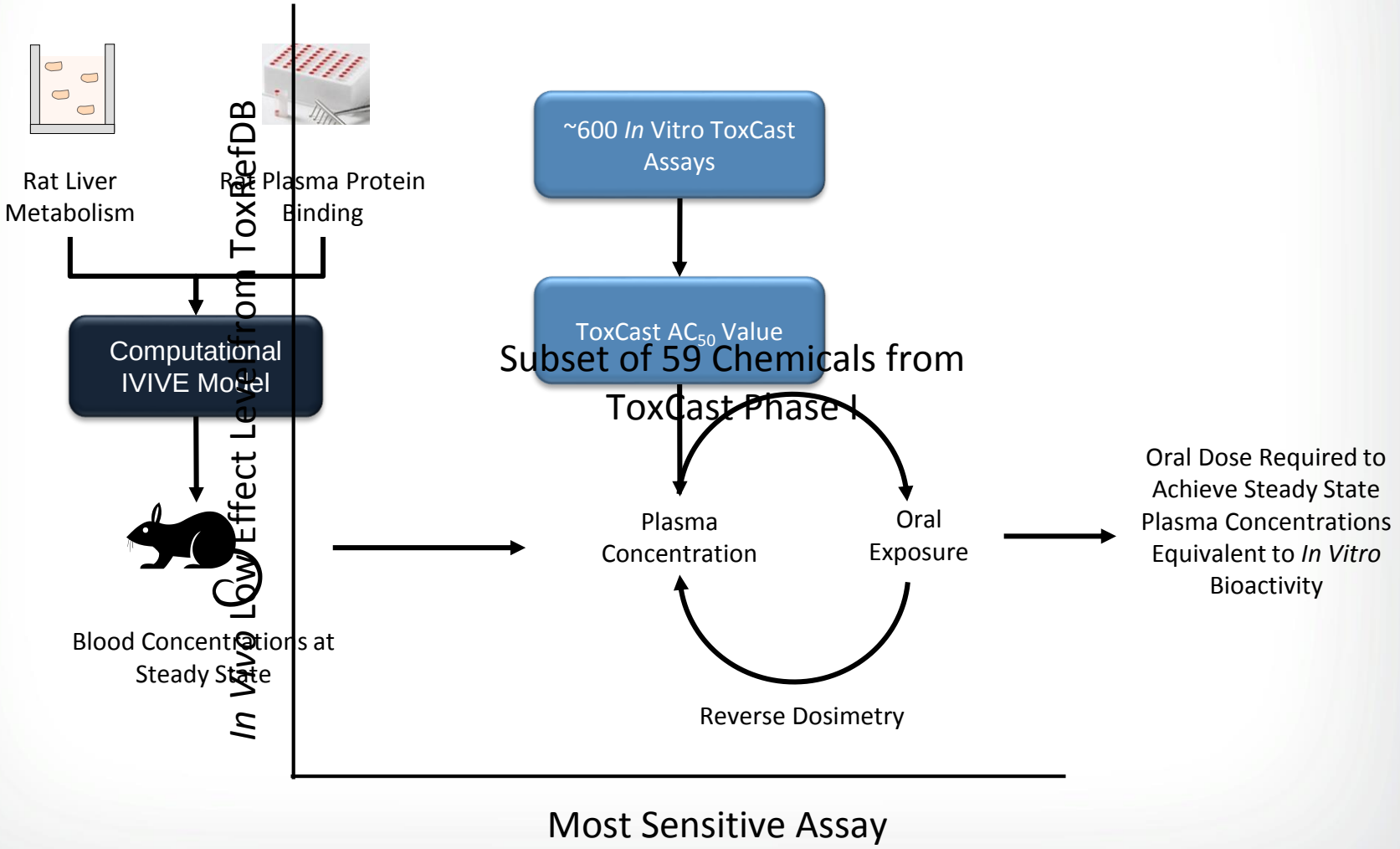
- Potentially related to non-selective interactions with multiple biological targets



Could we identify region of no biological perturbation and use that to define a point of departure?

Relatively narrow concentration range going from no biological activity to lots of activity/cytotoxicity

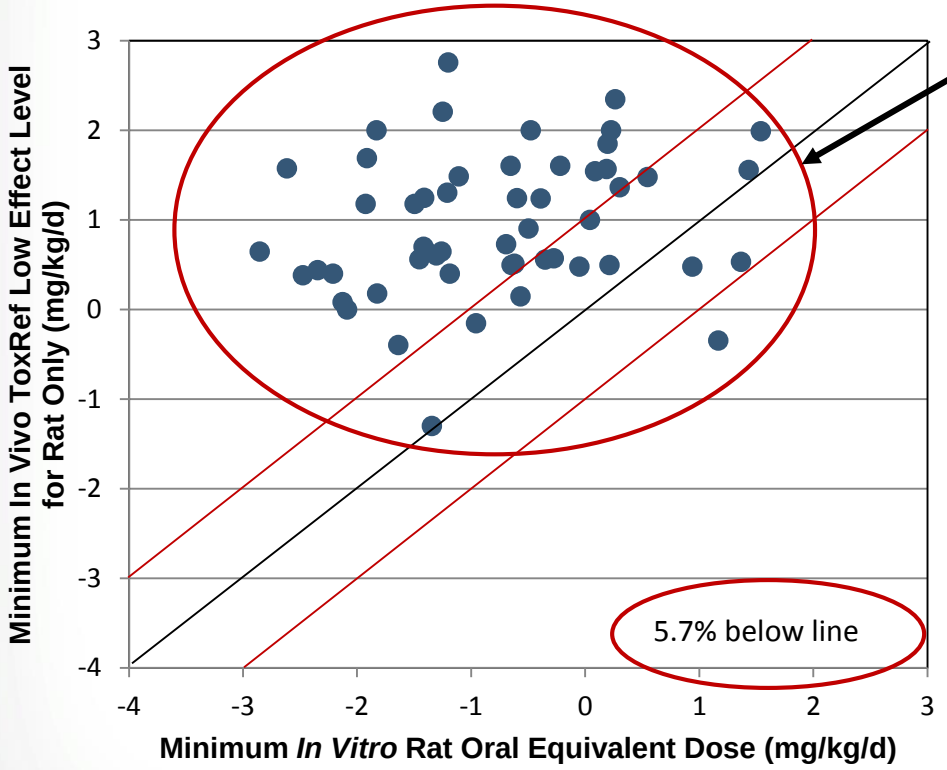
Evaluating ToxCast Assays for Dose-Response of Nonselective Chemicals

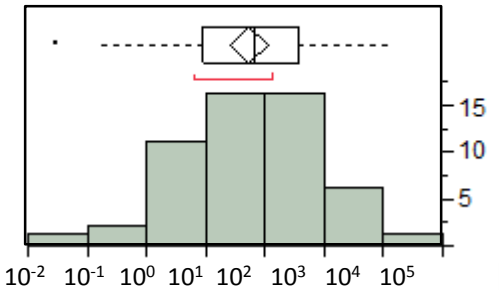


Wetmore *et al., Tox Sci.,* 2013

Region of No Biological Activity Provides a Conservative POD

Spanned 38 *In Vivo* Endpoints across Multiple Tissues, Organ Systems, and Study Types (Repro, Chronic, and Dev)





Log Ratio ToxRef Min LEL:ToxCast Min Oral Equivalent Dose

Distribution Summary Statistics		
Median	1.82	(66.07)
Upper Quartile	2.55	(354.81)
Lower Quartile	0.95	(8.91)

Wetmore *et al., Tox Sci.,* 2013

Integrating Human Dosimetry and Exposure

309 EPA ToxCast Phase I Chemicals

Human Liver Metabolism

Human Plasma Protein Binding

Dose (mg/kg/day)

Population-Based IVIVE Model

Oral Equivalent

Upper 95th Percentile Css
Among 100 Healthy Individuals of Both Sexes from 20 to 50 Yrs Old

~600 *In Vitro* ToxCast Assays

In Vitro Bioactivity

ToxCast AC_{50} Value

?

?

?

Plasma Concentration

Oral Exposure

Reverse Dosimetry

Chemical

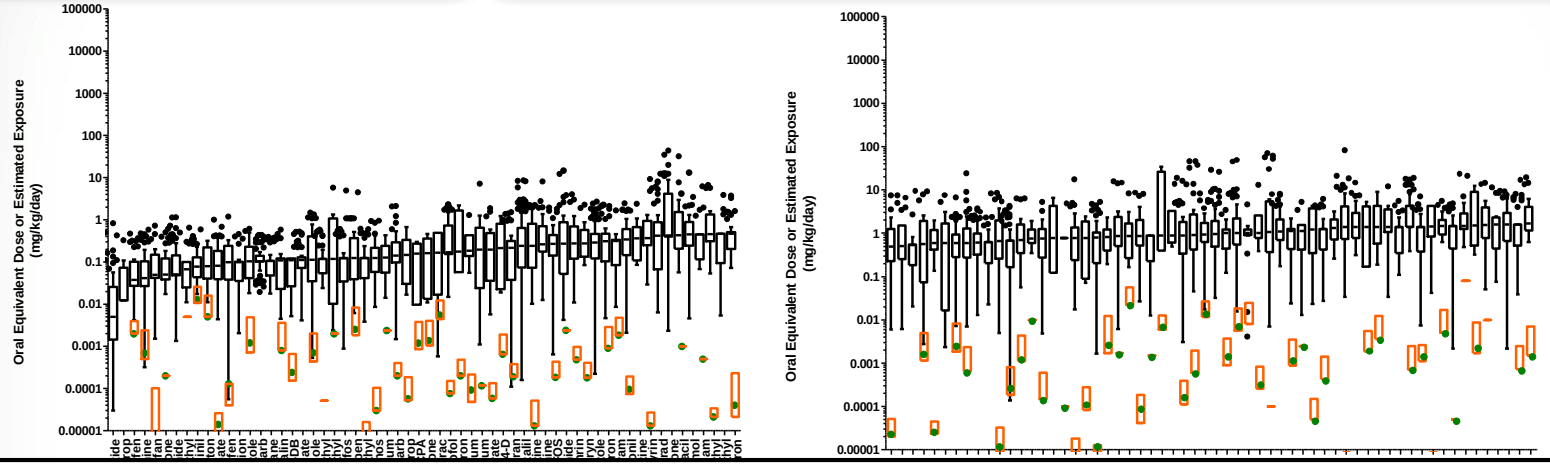
What are humans exposed to?

Least Sensitive Assay

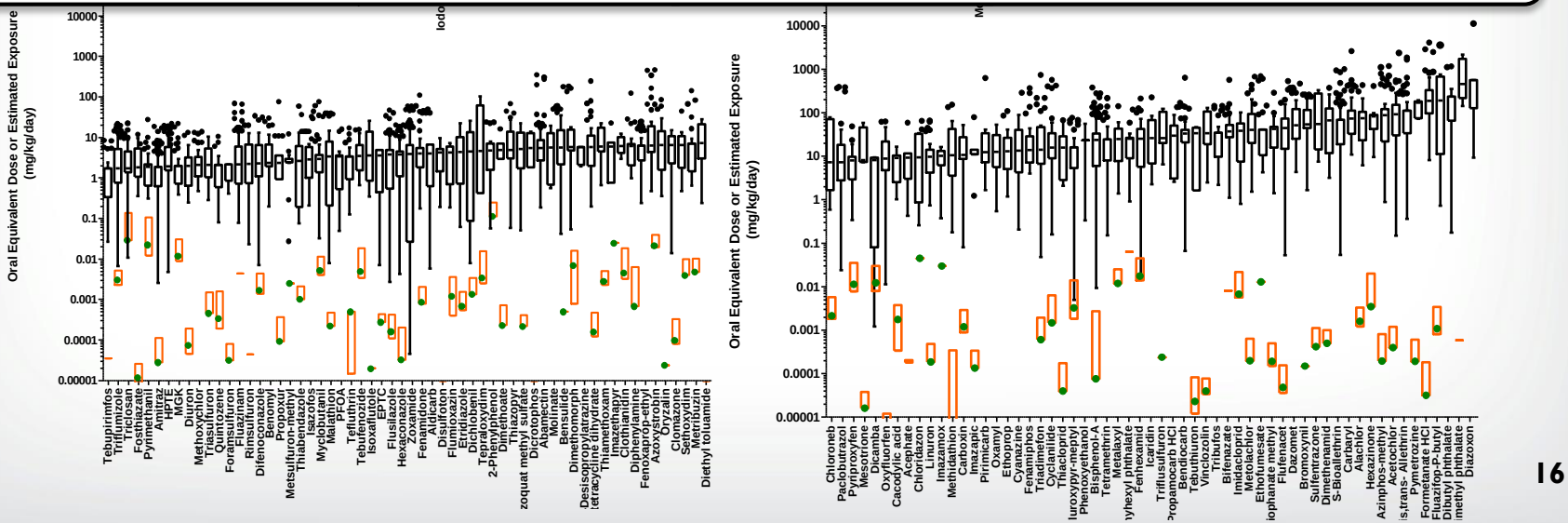
Oral Dose Required to Achieve Steady State Plasma Concentrations Equivalent to *In Vitro* Most Sensitive Assay Bioactivity

Rotroff *et al., Tox Sci.*, 2010
Wetmore *et al., Tox Sci.*, 2012

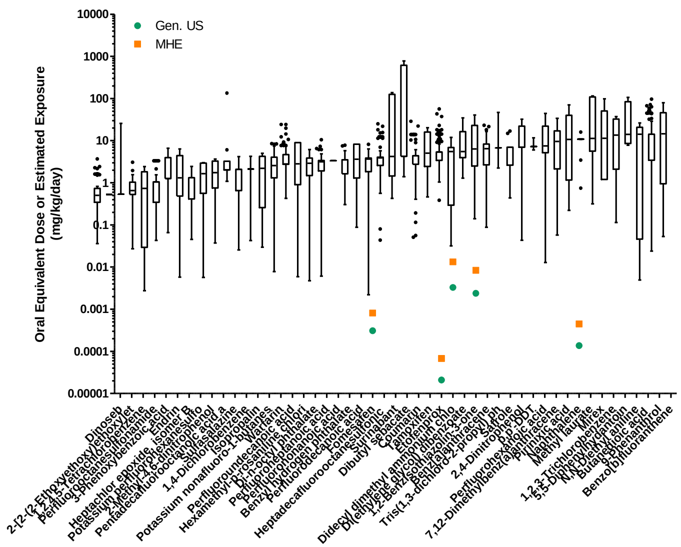
Margins-of-Exposure Can Identify Chemicals of Greatest Concern

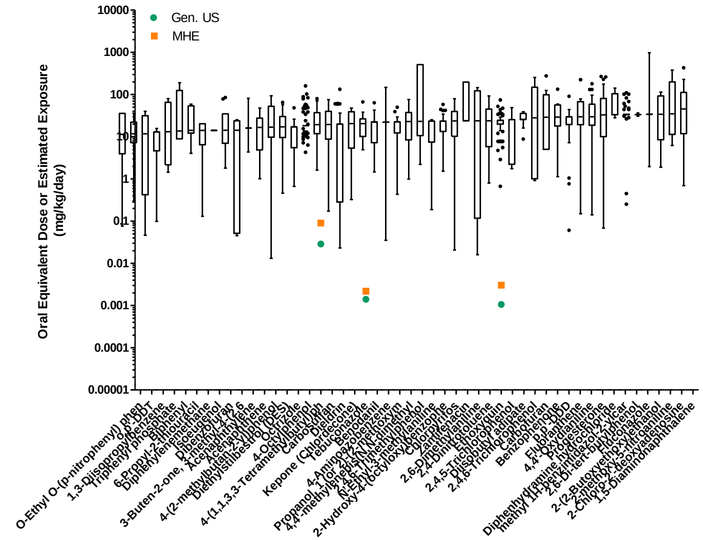


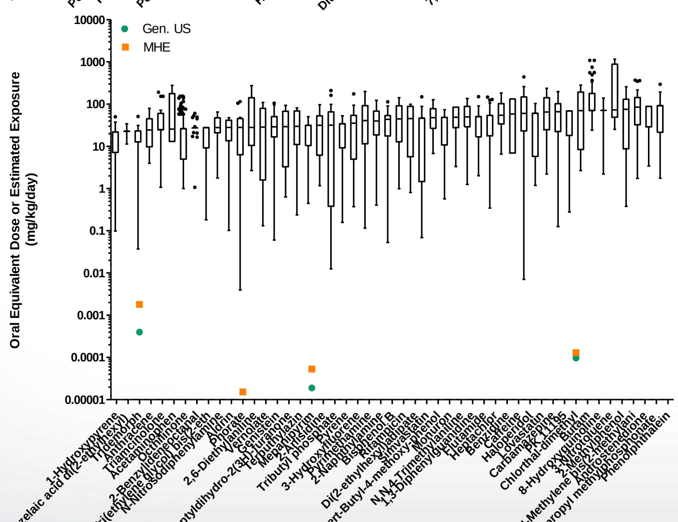
A total of 9.9% of the chemicals tested in ToxCast program have *in vitro* biological activity at oral equivalent doses that overlap the most highly exposed subpopulation.

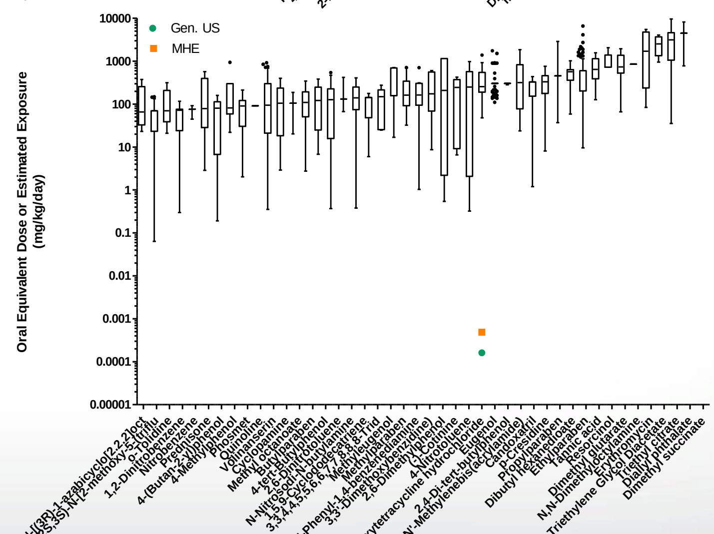


Then Came ToxCast Phase II…









High-Throughput Exposure Models Filling in Critical Data Gaps

(Bio) Monitoring

Dataset 1

Dataset 2

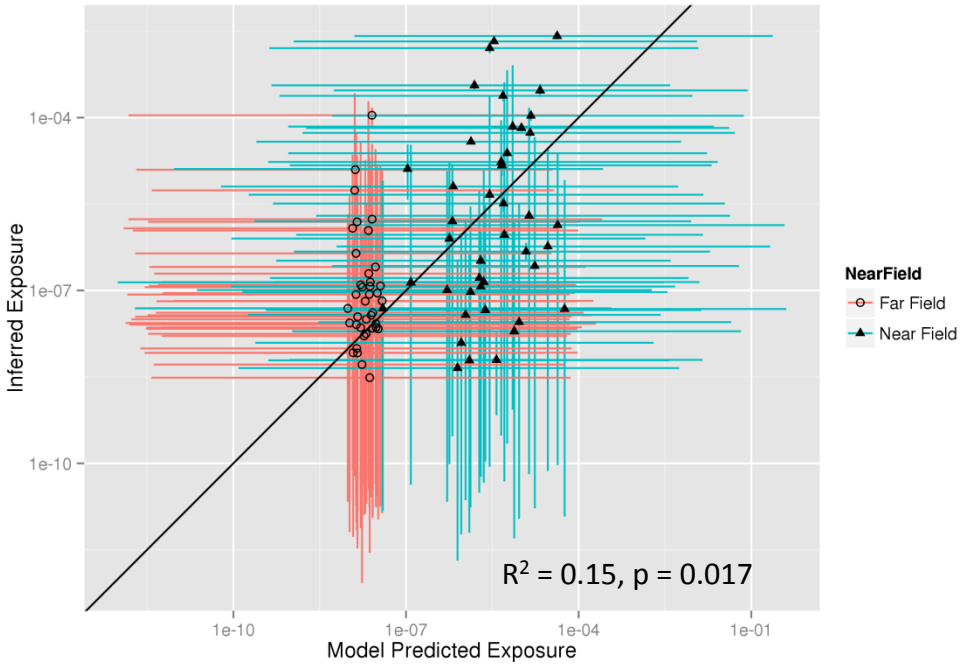
...

Model 1

Model 2

...

Established Fate and Transport Models

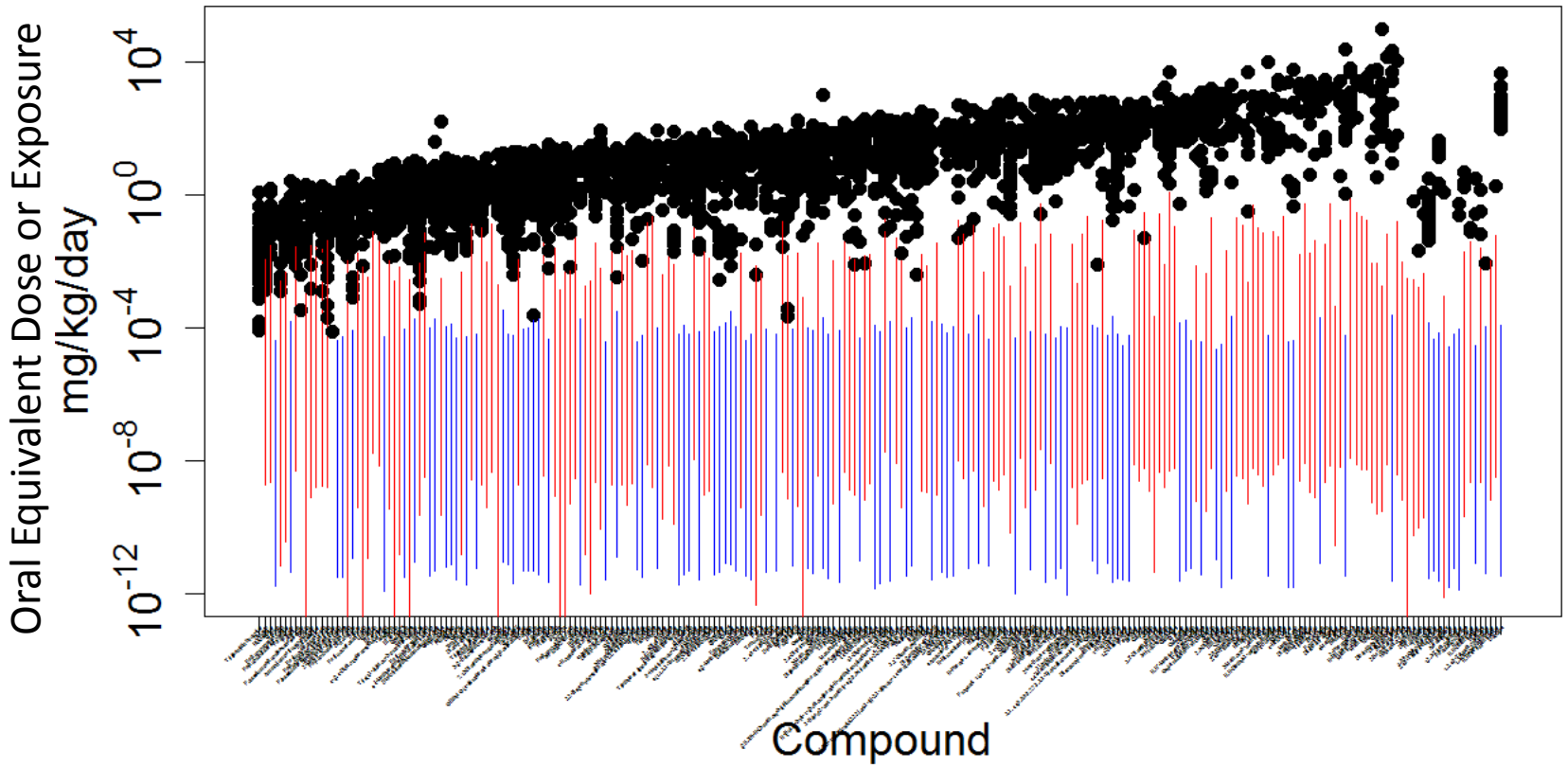


Inferred Exposures

Predicted Exposures

Wambaugh *et al., Env Sci Technol.,* 2013

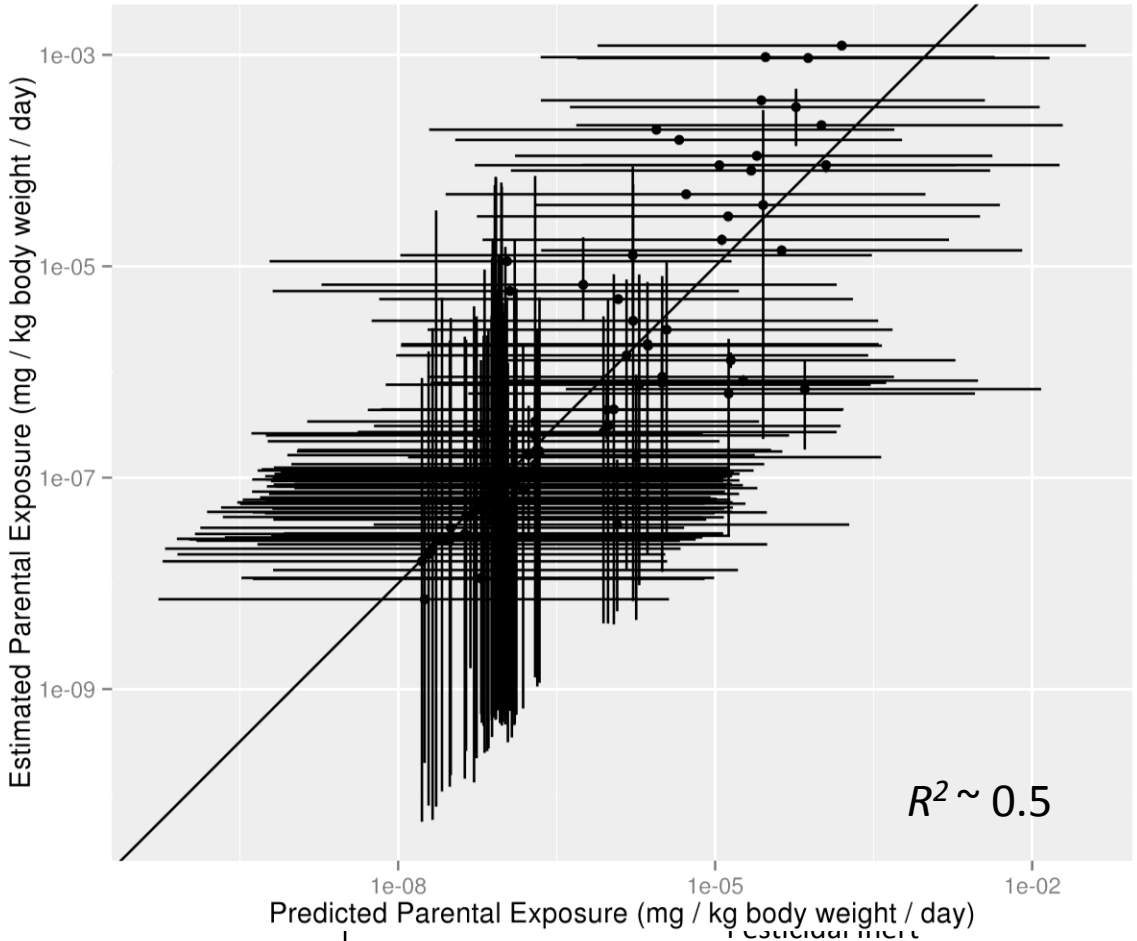
High-Throughput Exposure Models Filling in Critical Data Gaps



Oral Equivalent Doses Provided by Barbara Wetmore

Refining Exposure Model With Additional Chemical Use Categories

12 Chemical Use
Antimicrob
Chemical Industri
Consume
Dyes and Col
Fertilizer
Food Addi
Fragranc
Herbicid
Personal Care P
Pesticide
Petrochemi
Other

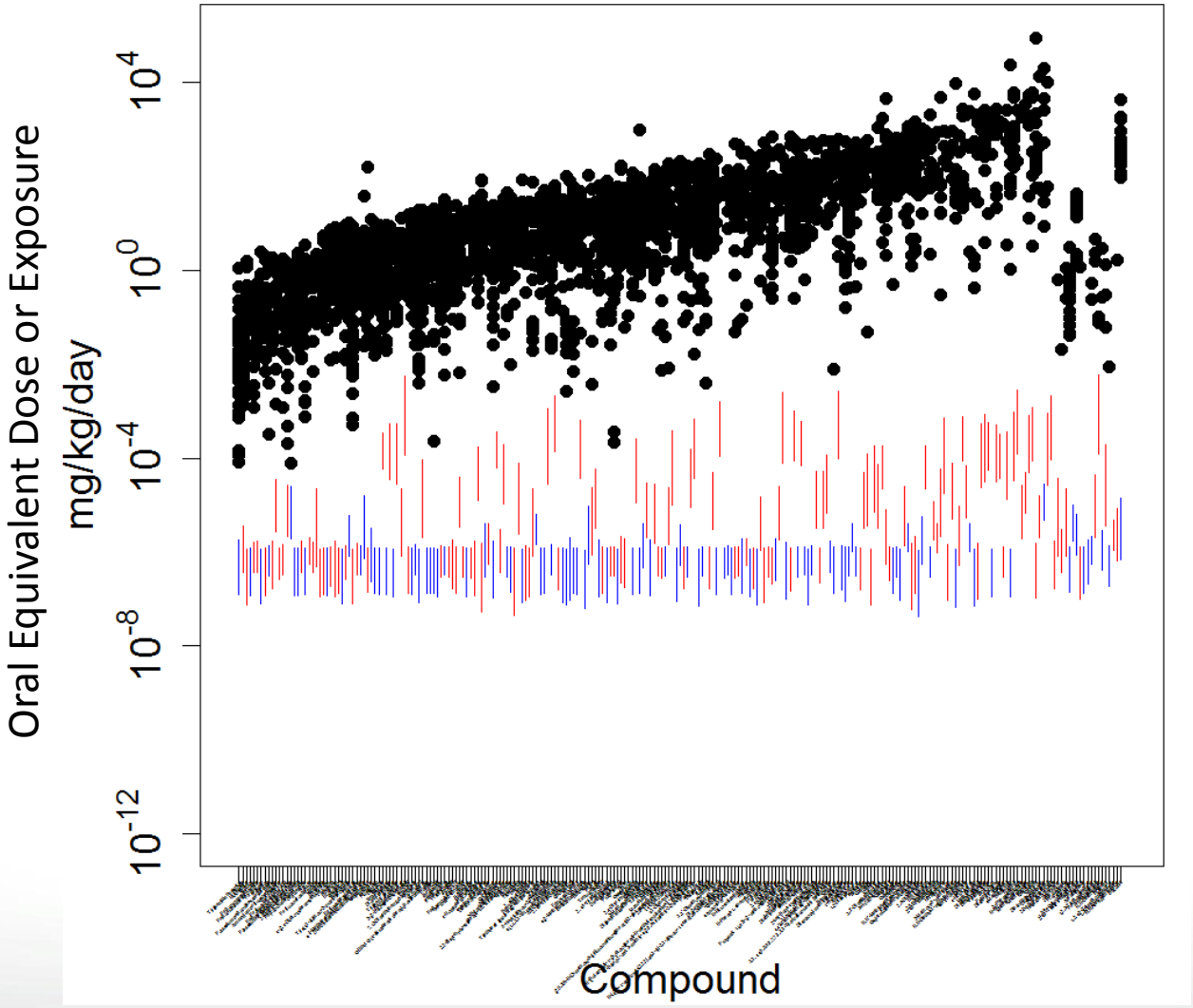


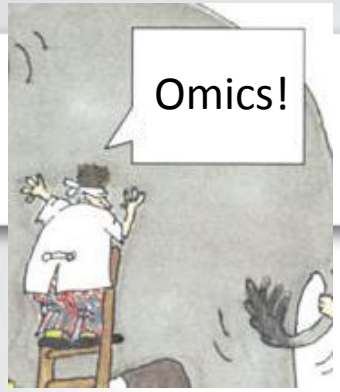
ry 12

ry 12

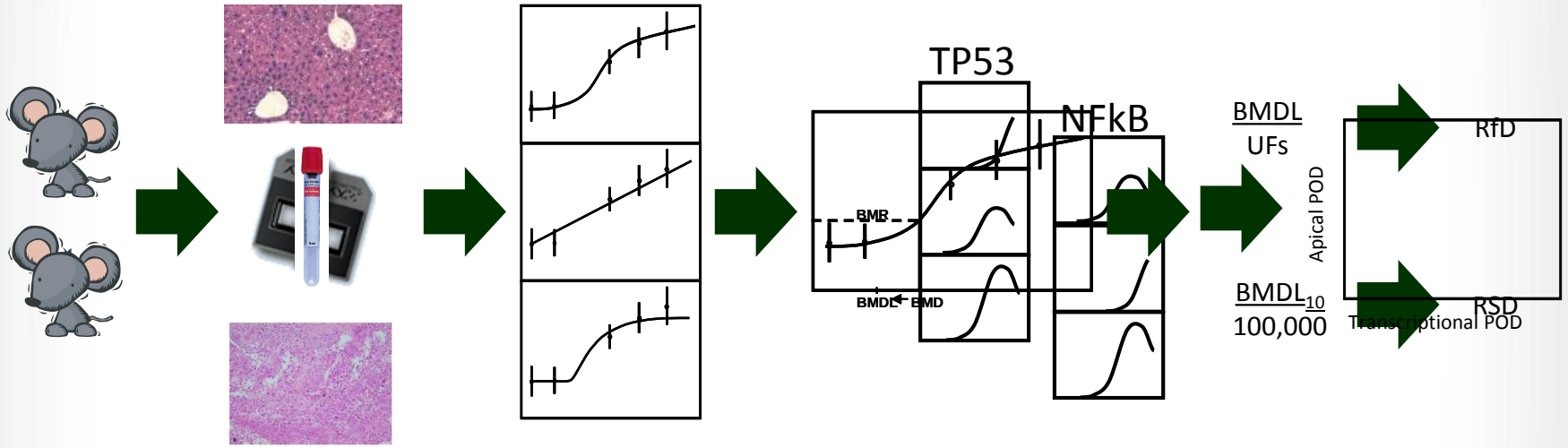
Production Volume

Second Generation High-Throughput Exposure Models Have Less Uncertainty





What About 'Omics?



Subchronic and Chronic Animal Studies

Most Sensitive Pathological Response Considered Adverse

Transcriptional Response

Fit Data with Statistical Models

Identify Point of Departure (i.e., BMDL)

Identify Genes By Pathway

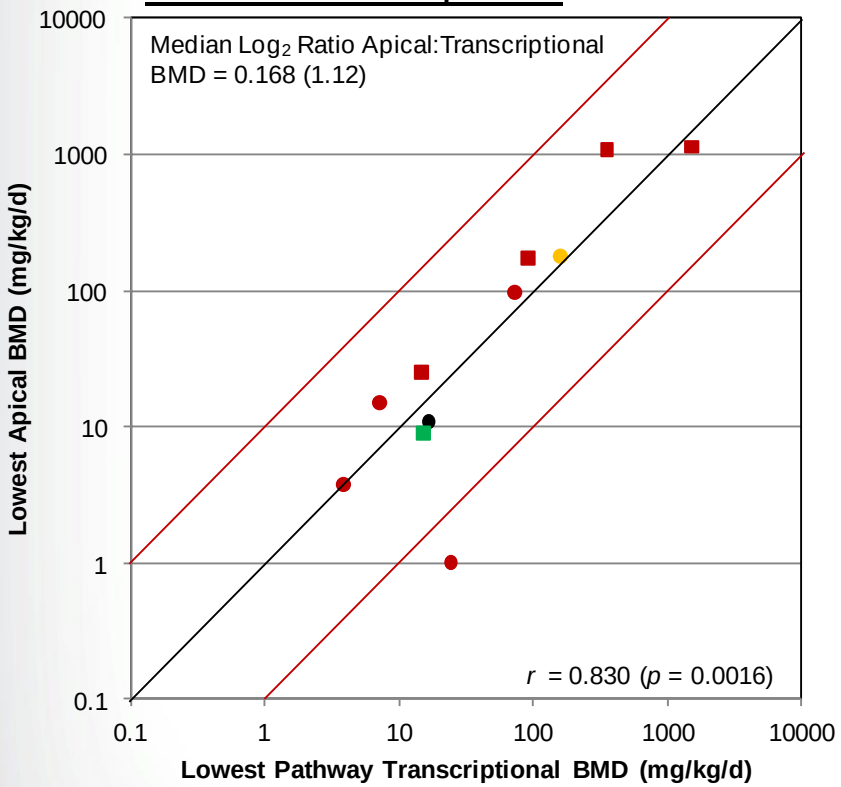
Derive Safe Exposure Level

Compare *In Vivo* Apical and Transcriptional PODs

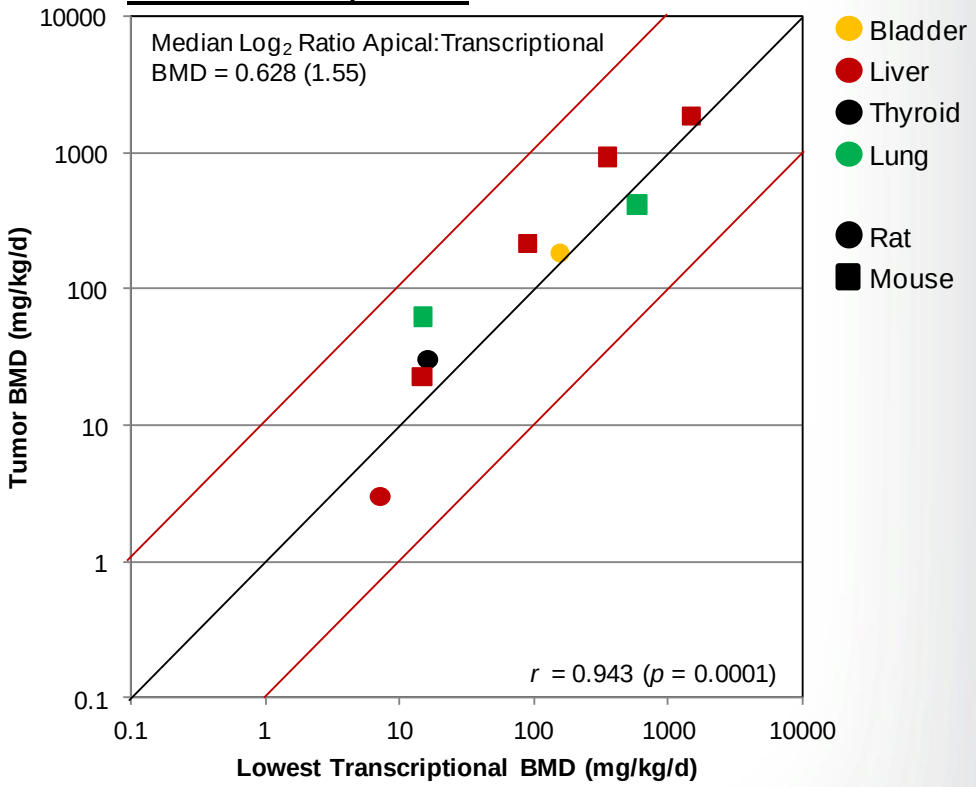
Thomas *et al., Tox Sci.*, 2011
Thomas *et al., Mut Res.*, 2012
Thomas *et al., Tox Sci.*, 2013

In Vivo Transcriptomics Provides an Accurate Estimate of the POD

Noncancer Endpoints

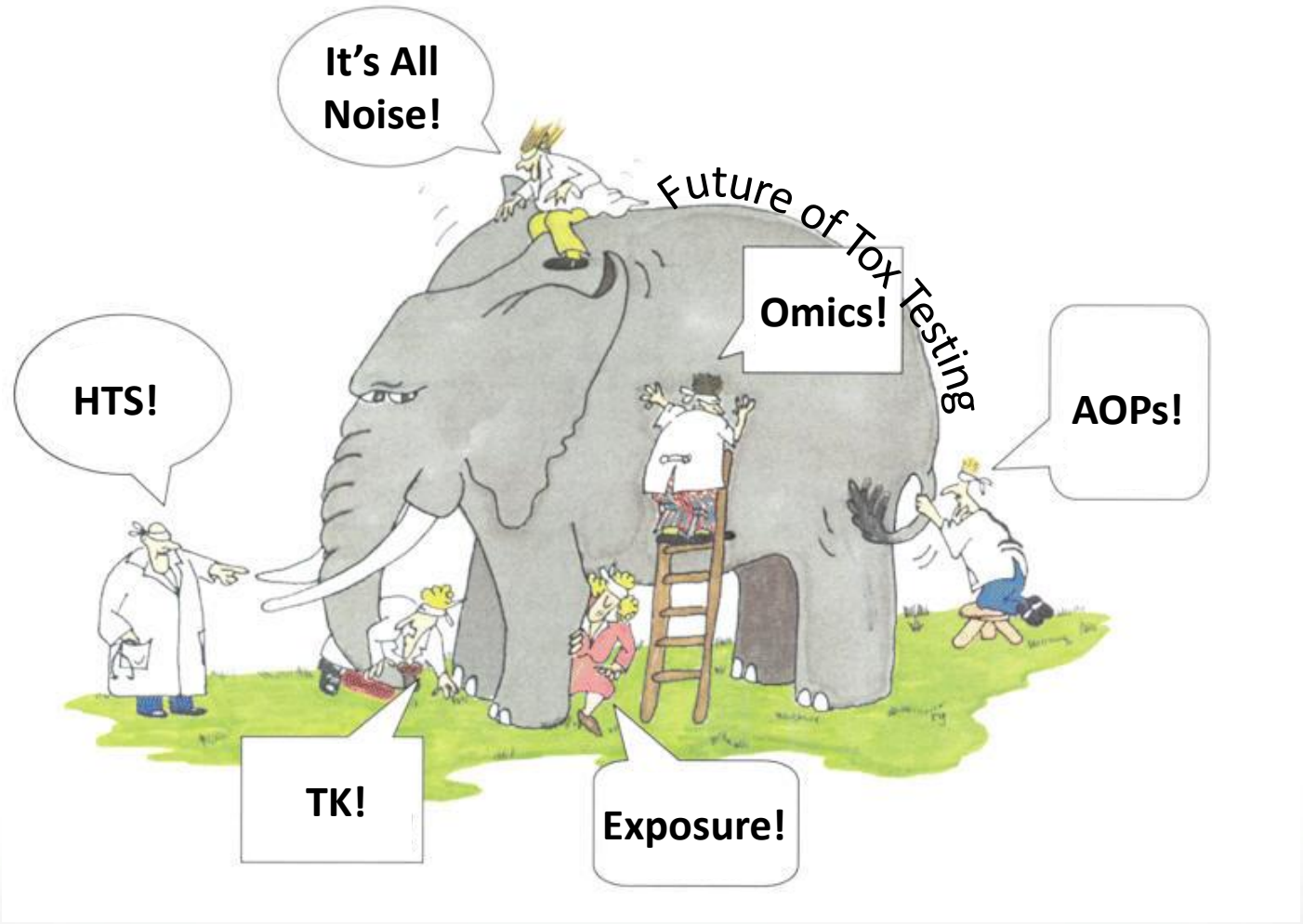


Cancer Endpoints

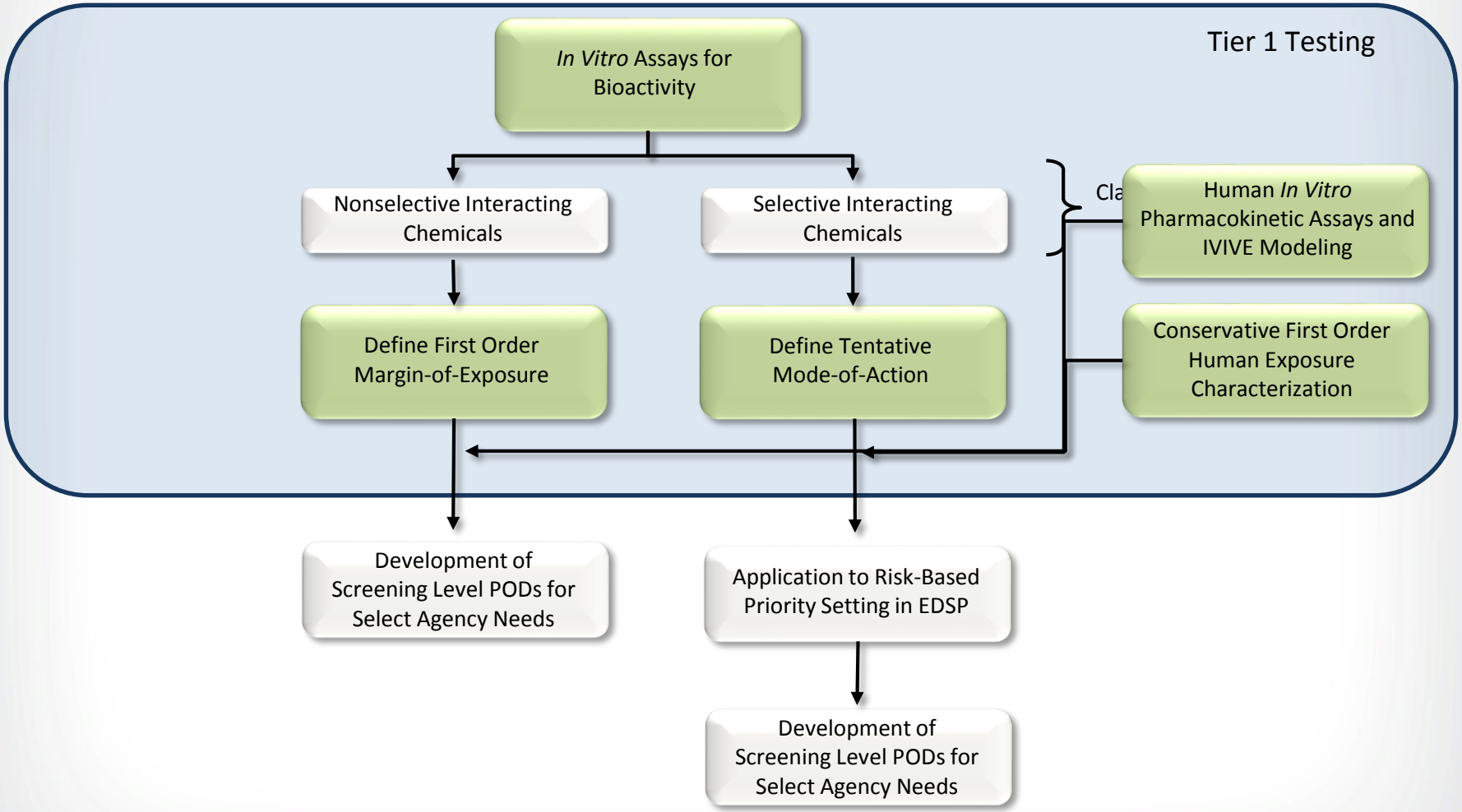


Thomas *et al., Tox Sci.,* 2013

Piecing Together What That Elephant May Look Like…

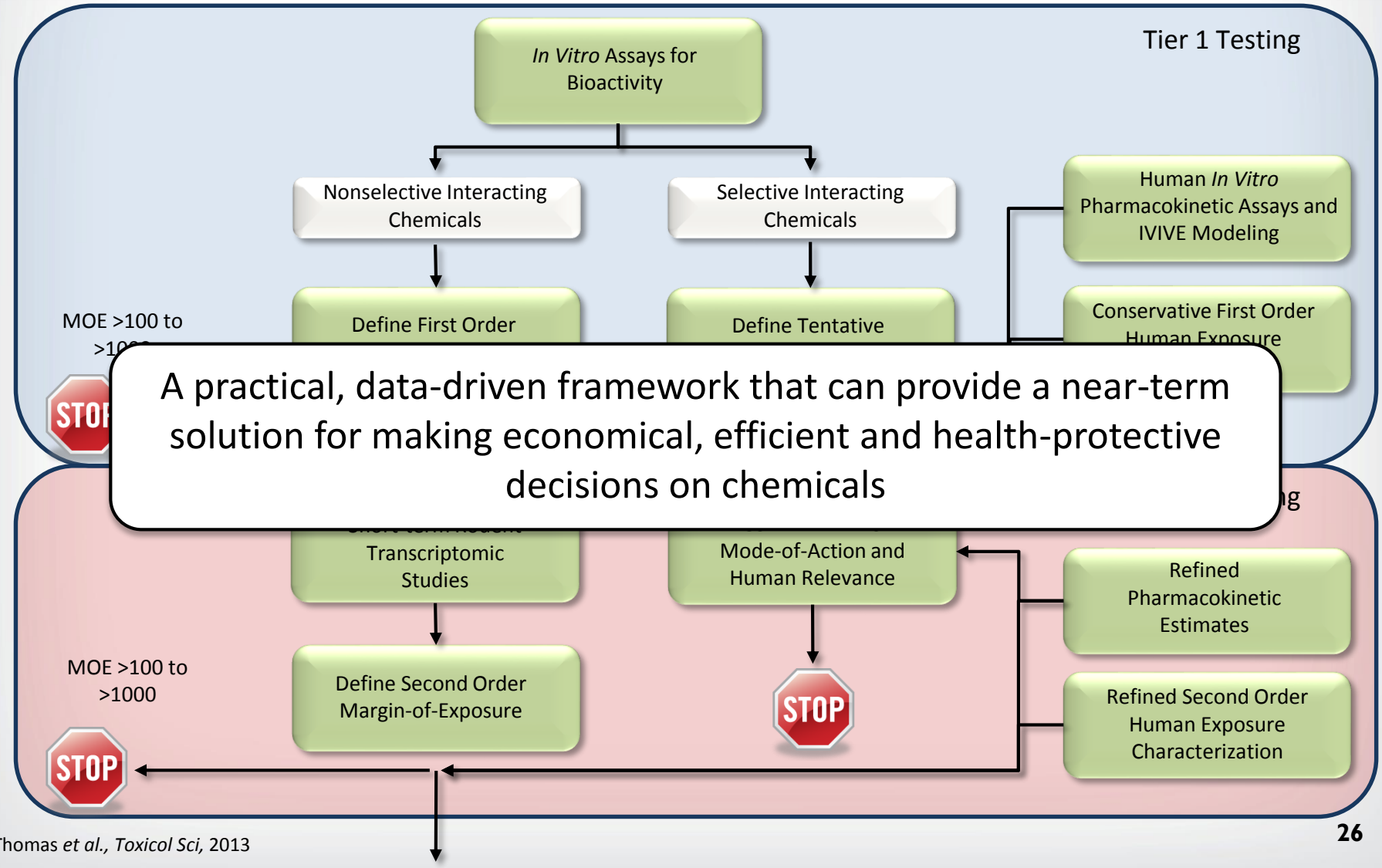


An Initial View of the Elephant



Thomas *et al., Toxicol Sci,* 2013

An Initial View of the Elephant



Thomas *et al., Toxicol Sci,* 2013

Acknowledgements

Tox21 Colleagues:
- NTP Crew
- FDA Collaborators
- NCGC Collaborators