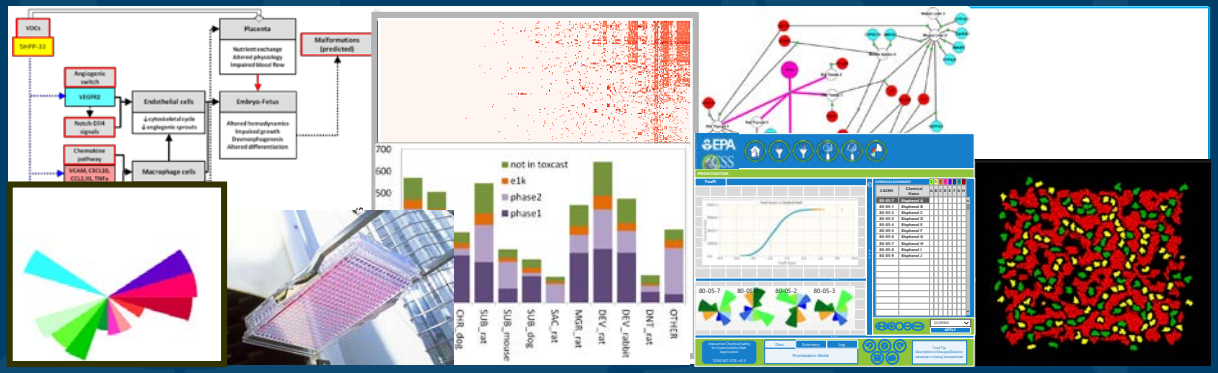
United States
Environmental Protection
Agency

Application of High-Throughput In Vitro Assays for Risk-Based Chemical Safety Decisions of Environmental and Industrial Chemicals



Society of Toxicology Annual Meeting
March 25, 2015

Rusty Thomas
Director
National Center for Computational Toxicology

Current System for Chemical Testing is Antiquated and Inefficient

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

Genotoxicity assays developed

Continuous breeding studies for reproductive toxicity

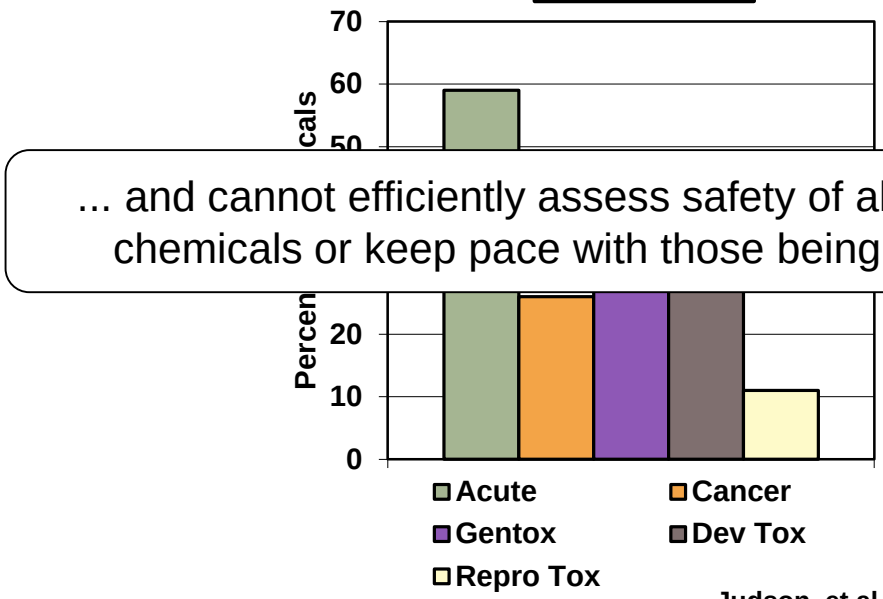
1920 | | 2010

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

Draize test introduced for eye irritants

Thalidomide led to testing of pharmaceuticals for developmental toxicity

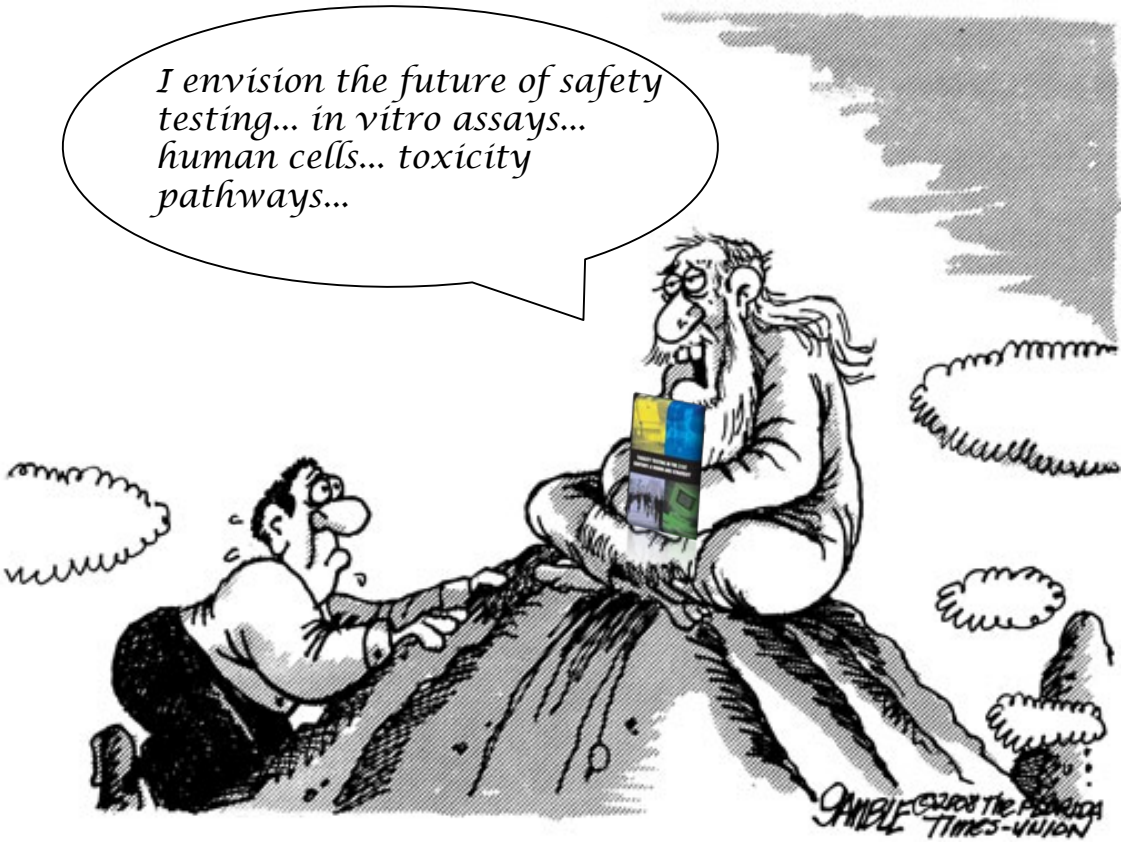
Rodent cancer bioassay introduced



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

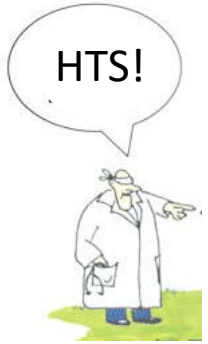
Judson, et al *EHP* (2010)

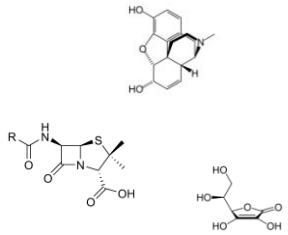
In 2007, NRC Transformed Toxicology with a Future View

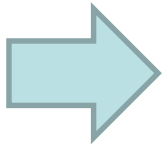


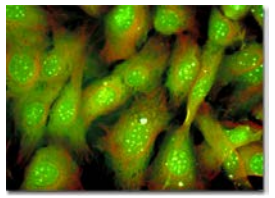
High Throughput Screening By Itself Leads is Incomplete

High-Throughput Screening is One Part of the Elephant…



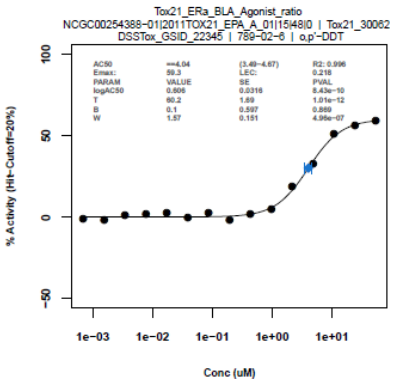






1,800 Chemicals in Concentration Response

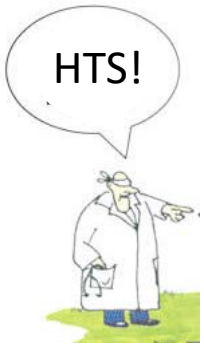
~300 Phase I
~700 Phase II
~800 E1K

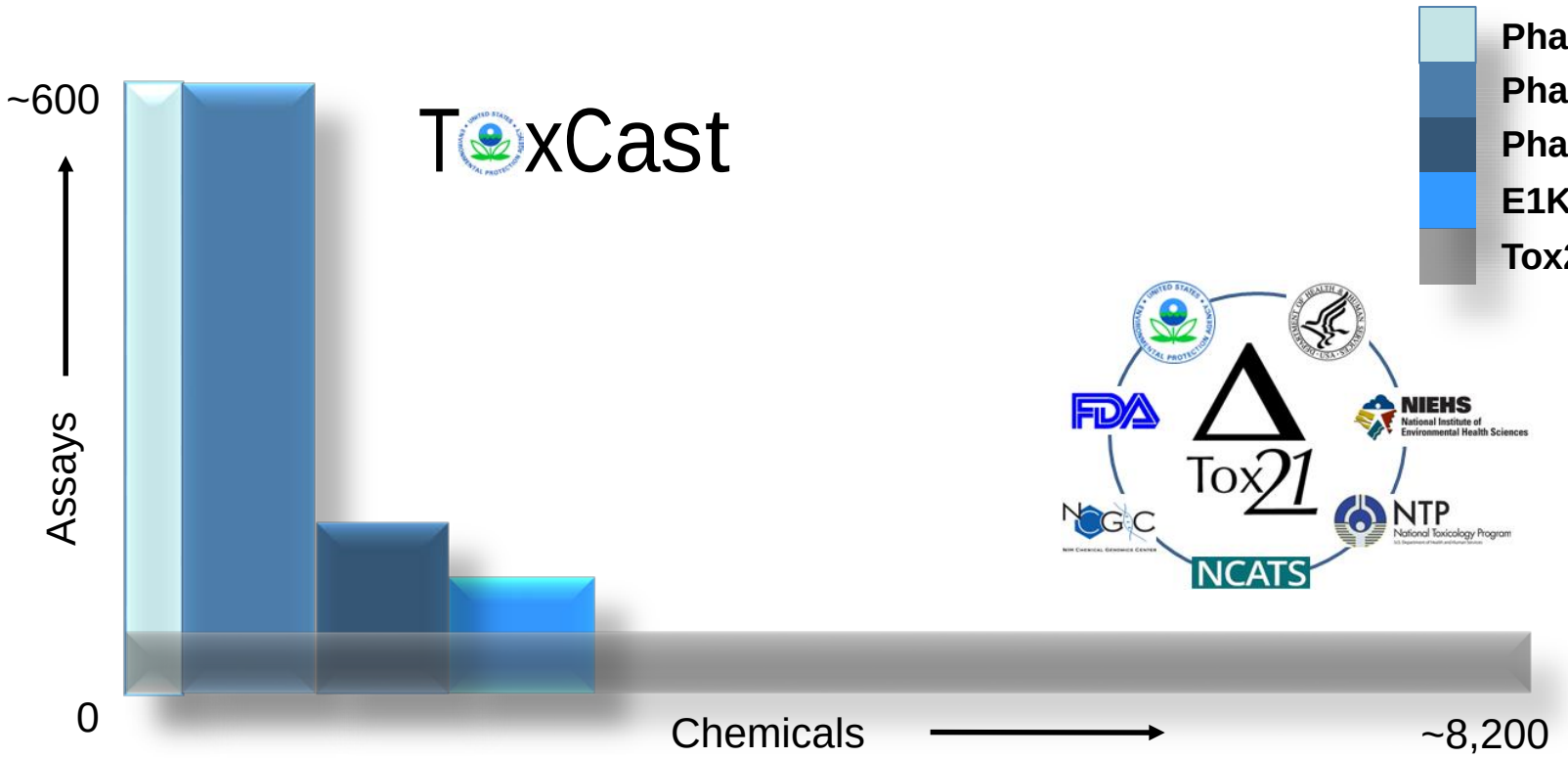


~700 Biochemical and Cell-based High-throughput Screening Assay Endpoints

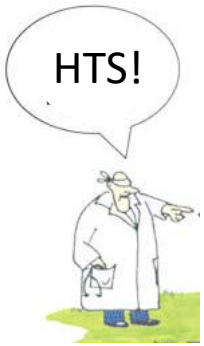
(327 genes and 293 pathways)

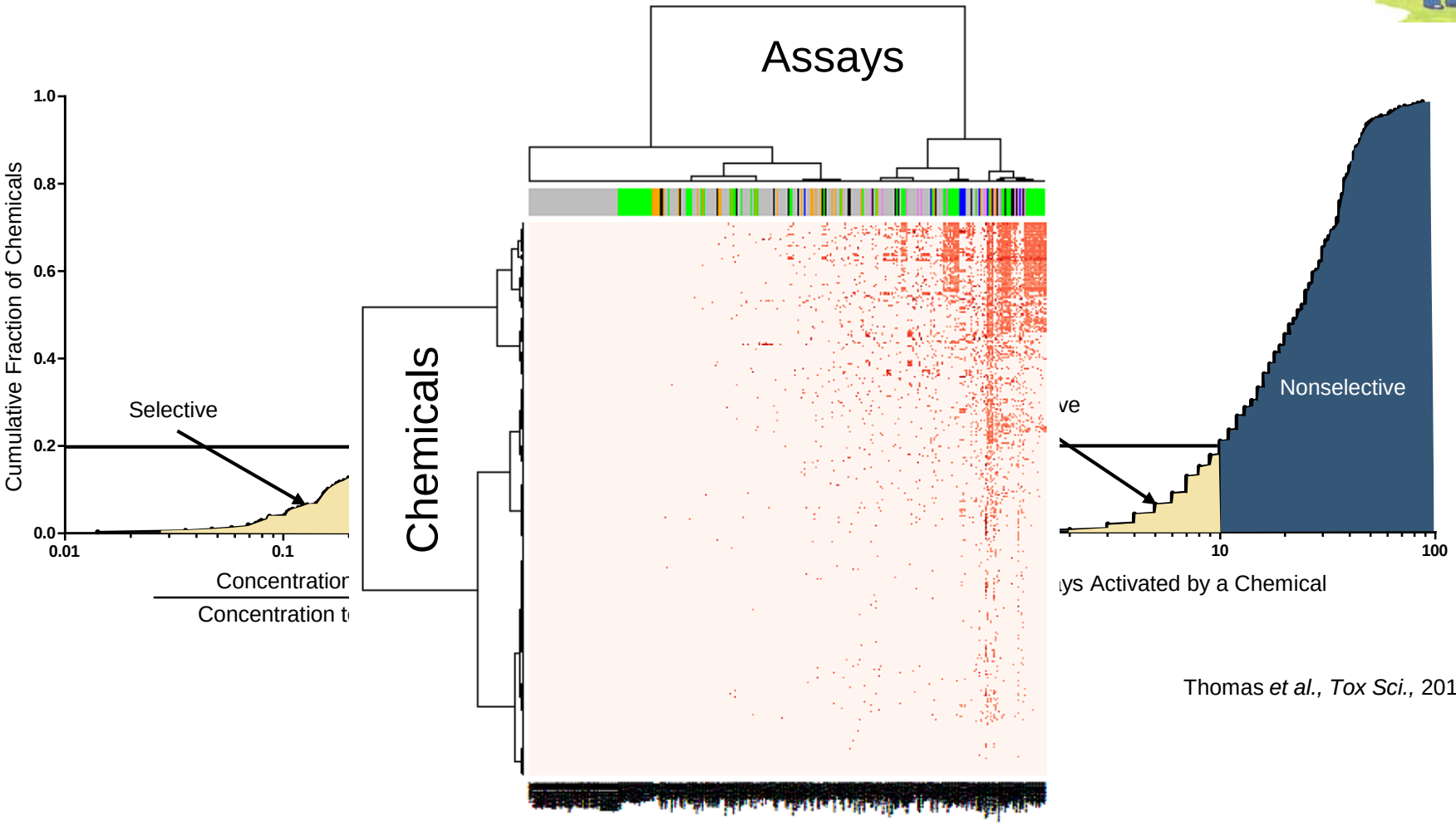
ToxCast and Tox21 Provide Complimentary Approaches





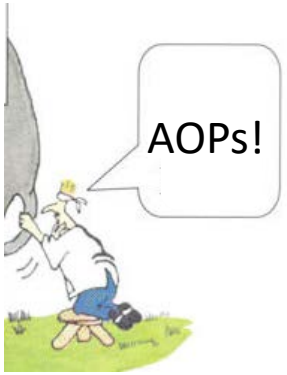
What Have We Learned From High-Throughput Screening?

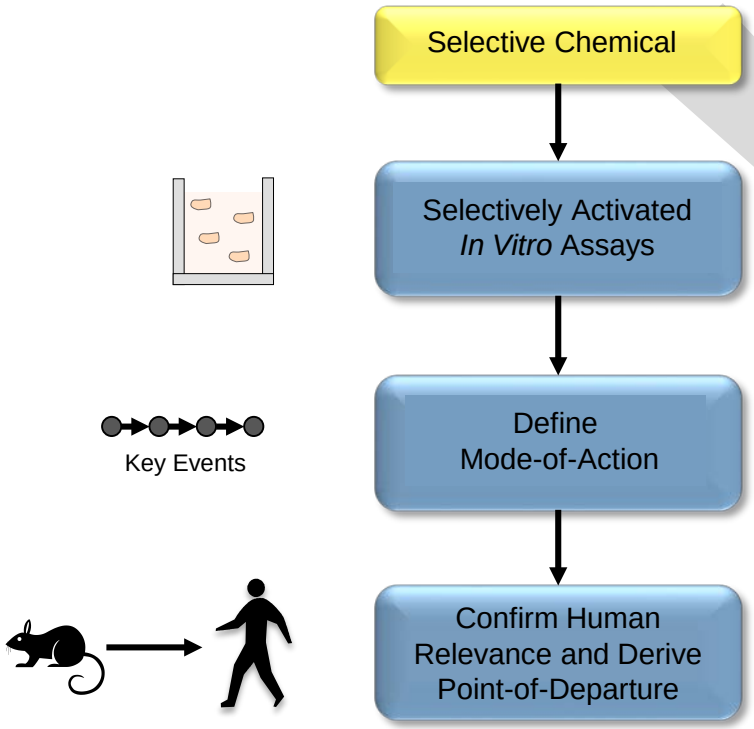


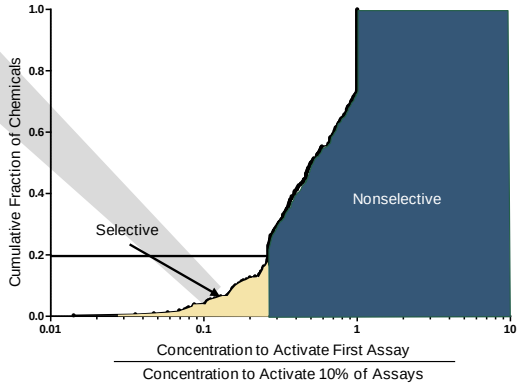


Thomas *et al., Tox Sci.*, 2013

In Vitro Assay Selectivity as a Starting Point for AOPs

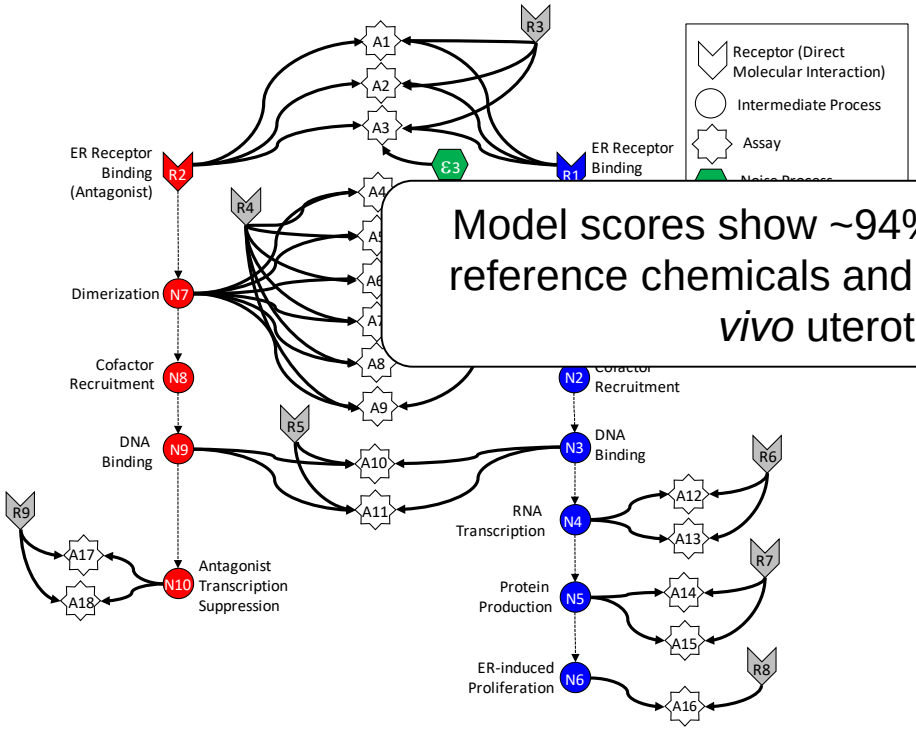




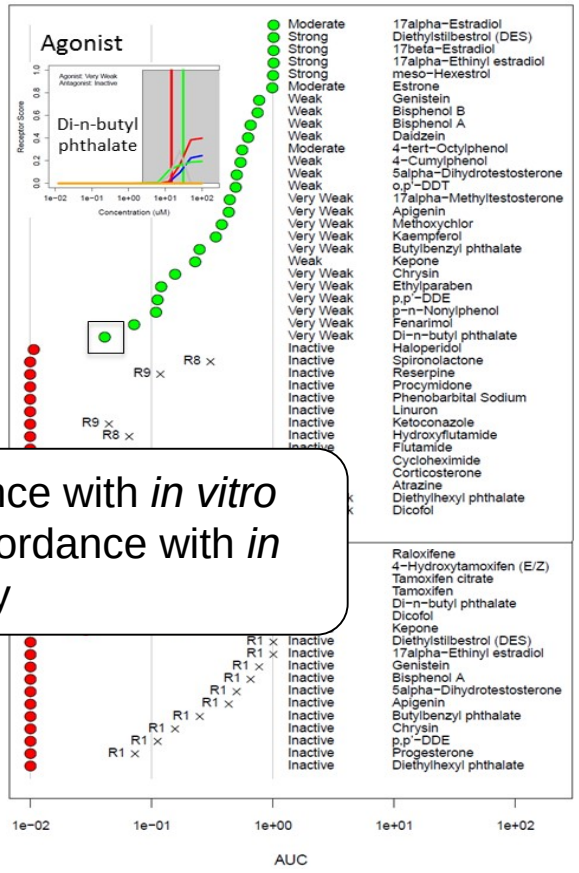


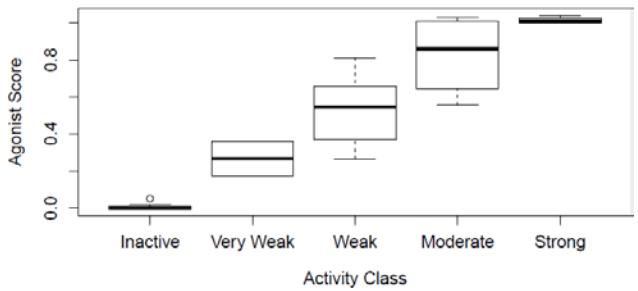
Computational Modeling to Integrate Upstream Events in AOPs

18 *In Vitro* Assays Measure ER-Related Activity

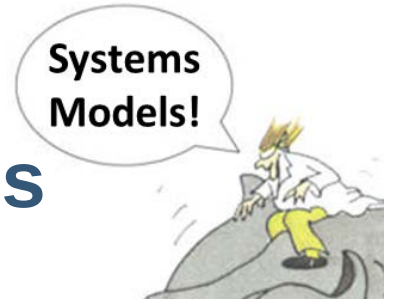


Model scores show ~94% concordance with *in vitro* reference chemicals and ~95% concordance with *in vivo* uterotrophic assay

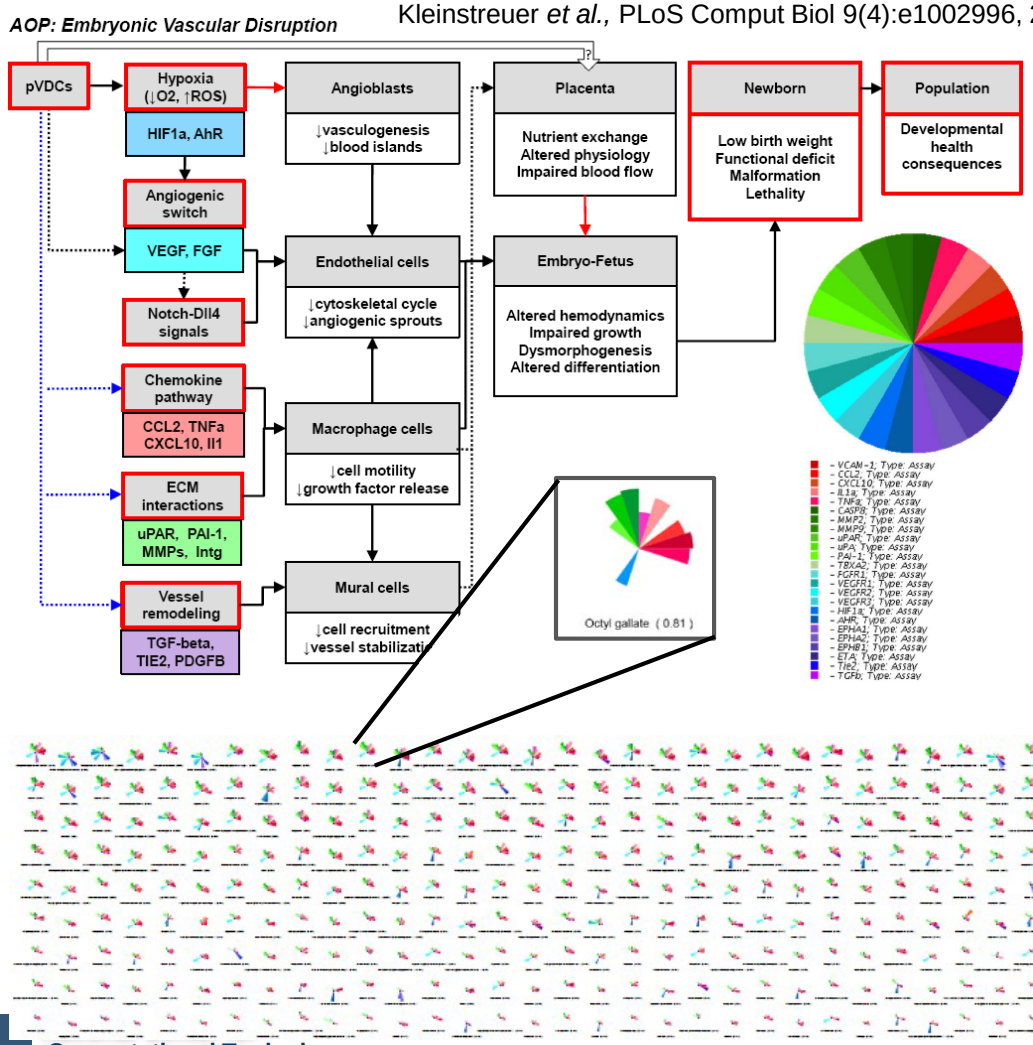




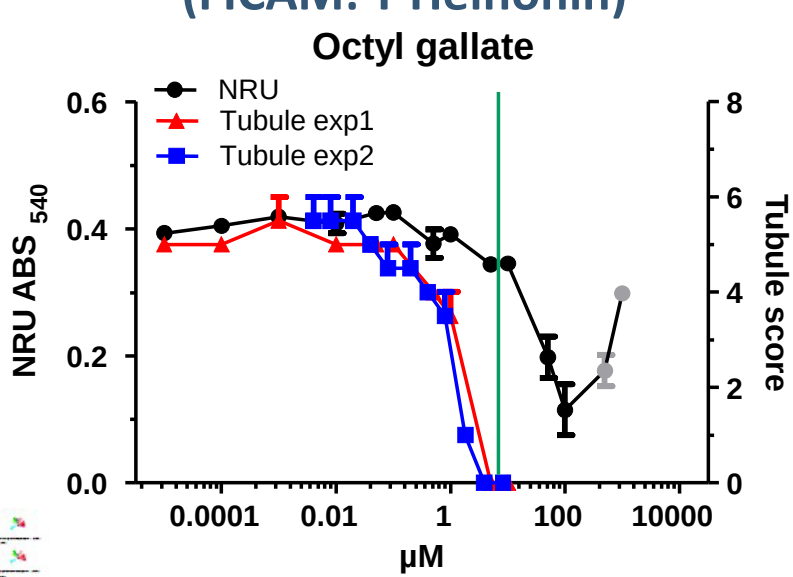
Integration of Assays for Multiple Key Events into AOPs



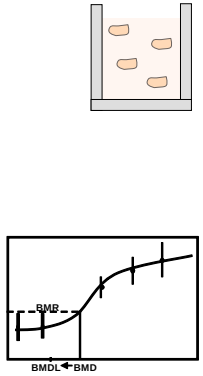
Kleinstreuer *et al.*, PLoS Comput Biol 9(4):e1002996, 2013



Human Tubulogenesis Assay (FICAM: T Heinonin)



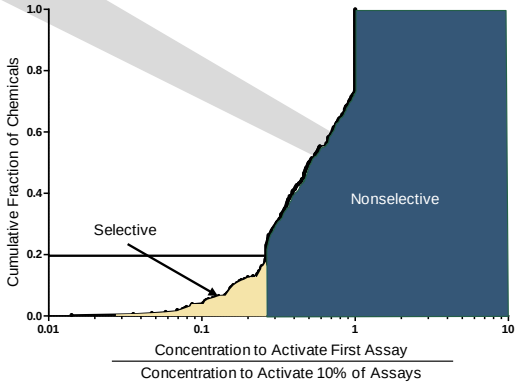
What About the Non-Selective Chemicals?



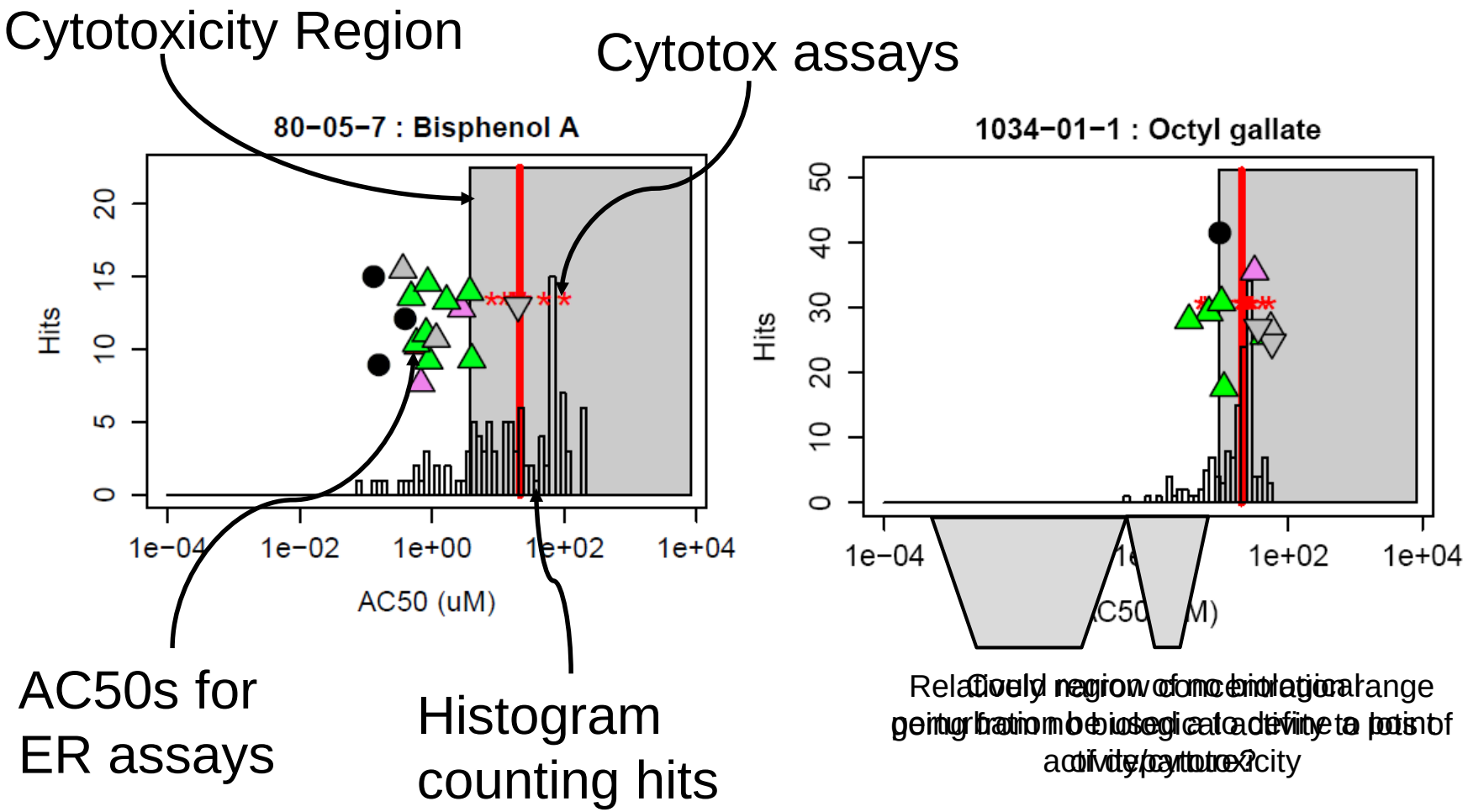
Nonselective Chemical

?????????

Define Point-of-Departure



Non-Selectivity Closely Aligned with Cytotoxicity



TK & Exposure Are Other Parts of the Elephant…

309 EPA ToxCast Phase I Chemicals

Human Liver Metabolism

Human Plasma Protein Binding

Population-Based IVIVE Model

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

Daily Exposure Dose (mg/kg/day)

~700 *In* Vitro ToxCast Assays

In Vitro Bioactivity

ToxCast AC_{50} Value

?

?

?

Plasma Concentration

Oral Exposure

Reverse Dosimetry

Chemical

Least Sensitive Assay

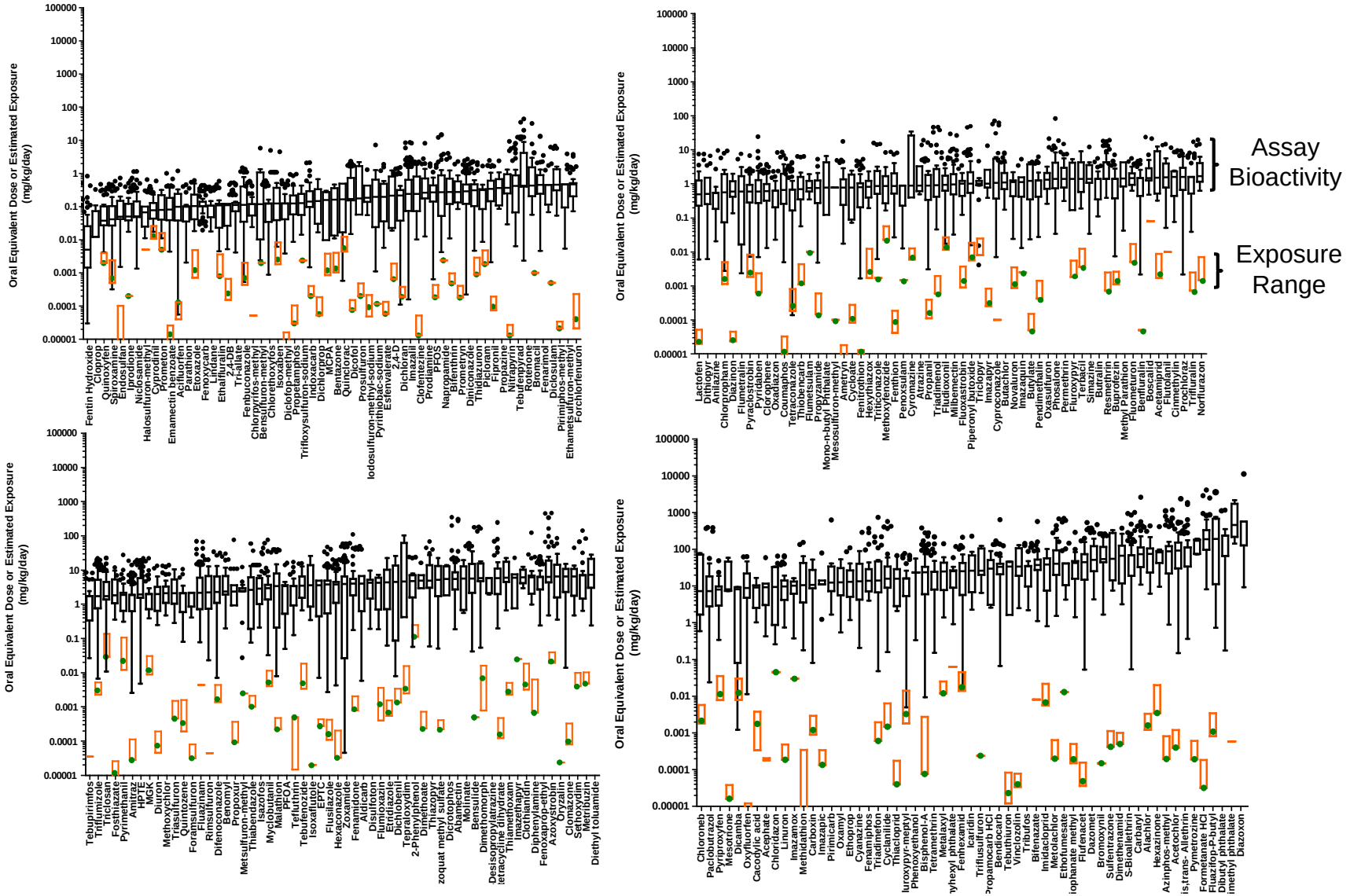
Oral Dose Required to Achieve Steady State Plasma Concentrations Equivalent to *In Vitro* Bioactivity

Most Sensitive Assay

What are humans exposed to?

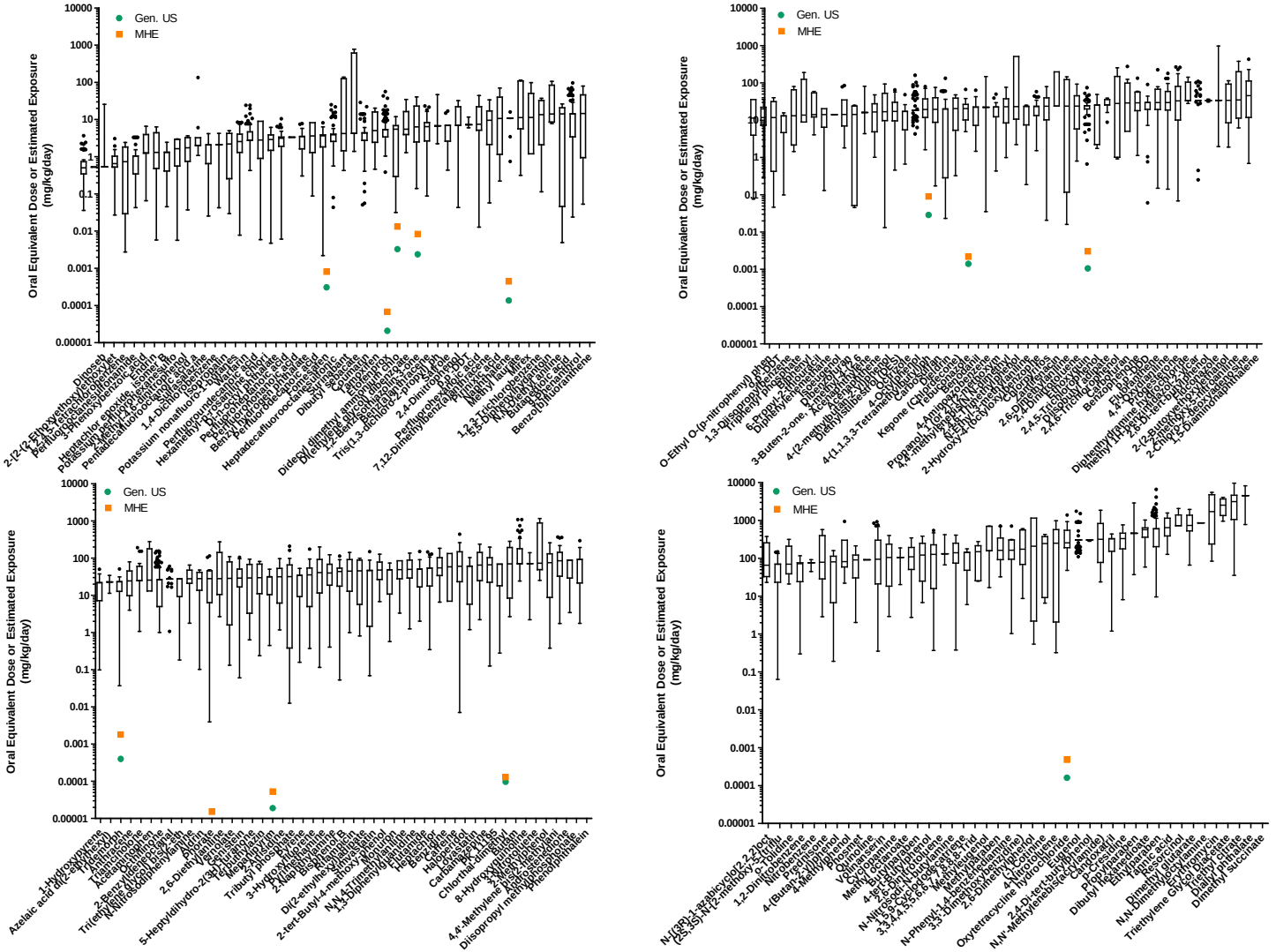
Rotroff *et al., Tox Sci.*, 2010
Wetmore *et al., Tox Sci.*, 2012

Comparing Assay Bioactivity with Exposure Provides Risk Context



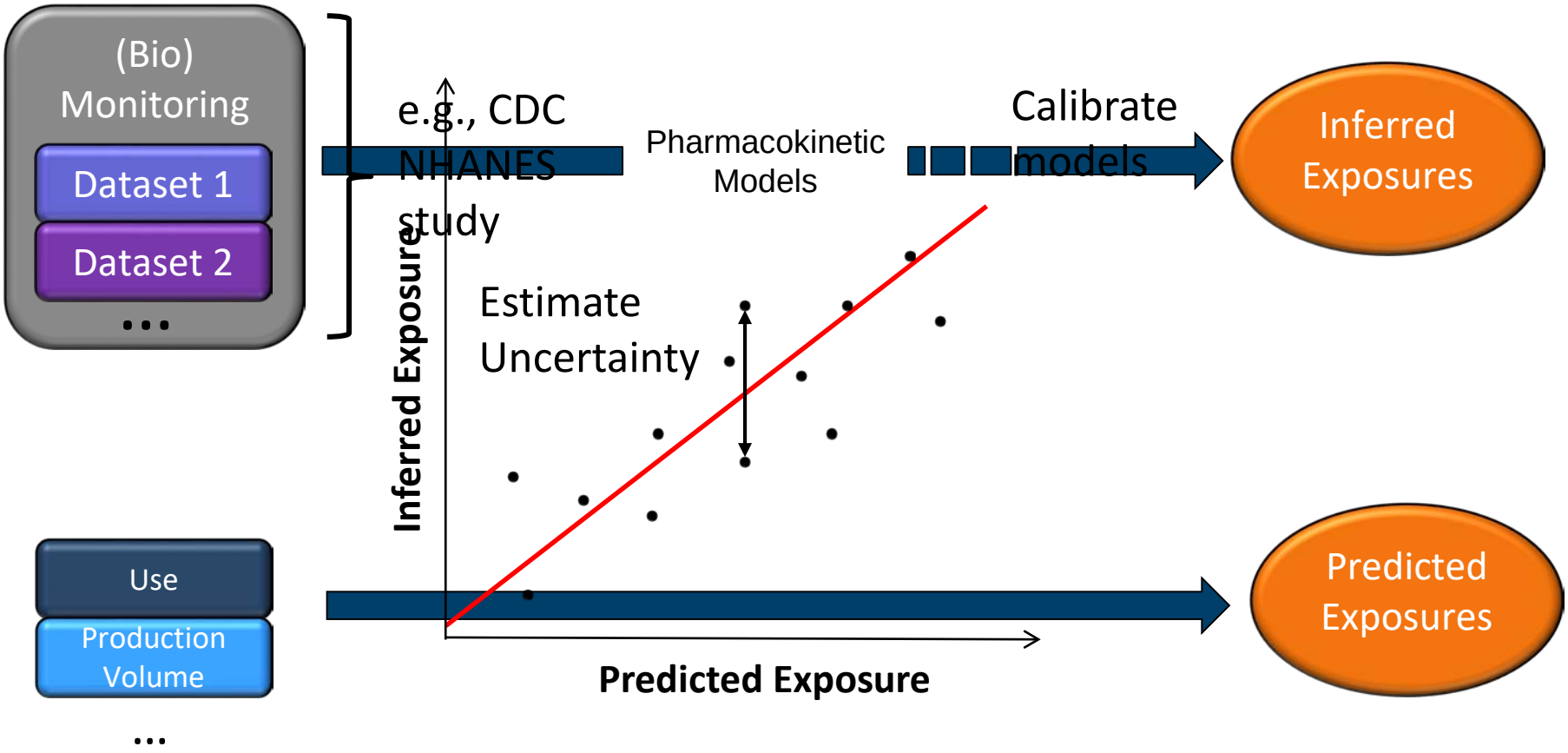
Wetmore *et al.*, *Tox Sci.*, 2012

Then Came ToxCast Phase II…



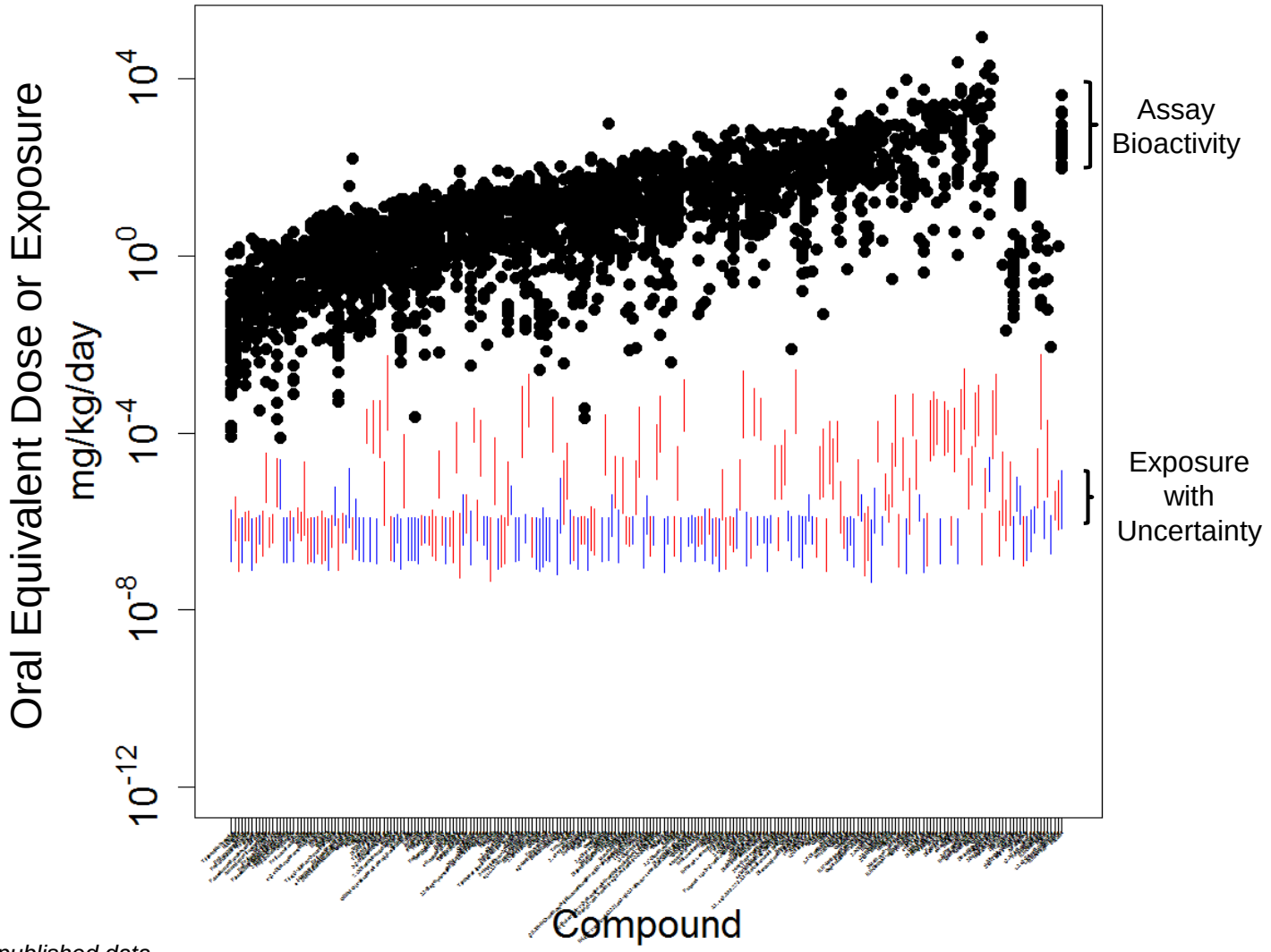
Courtesy Barbara Wetmore

High-Throughput Exposure Models Filling Critical Data Gaps



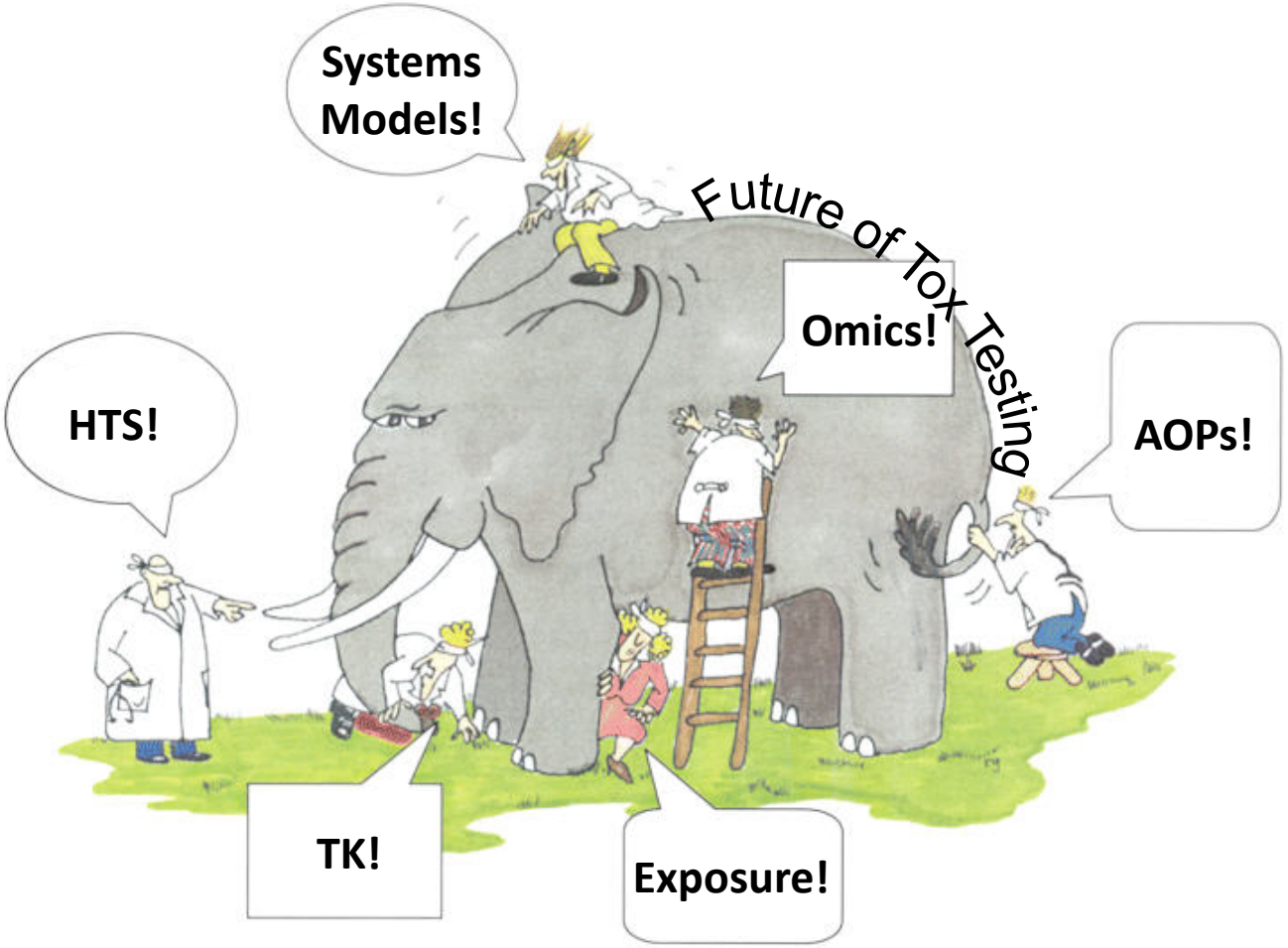
Wambaugh *et al., unpublished data*

Activity and Exposure Allow Robust Prioritization of Chemicals

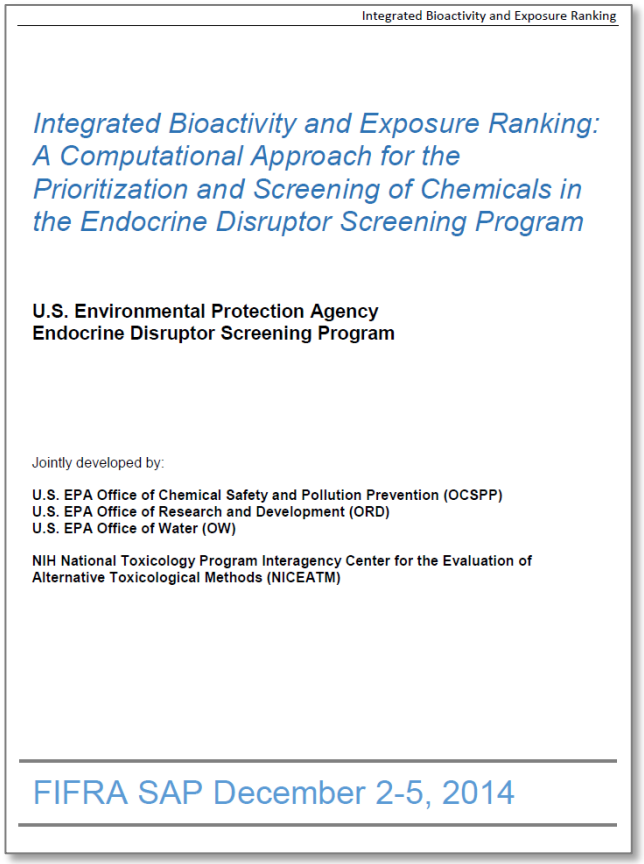


Wambaugh *et al., unpublished data*

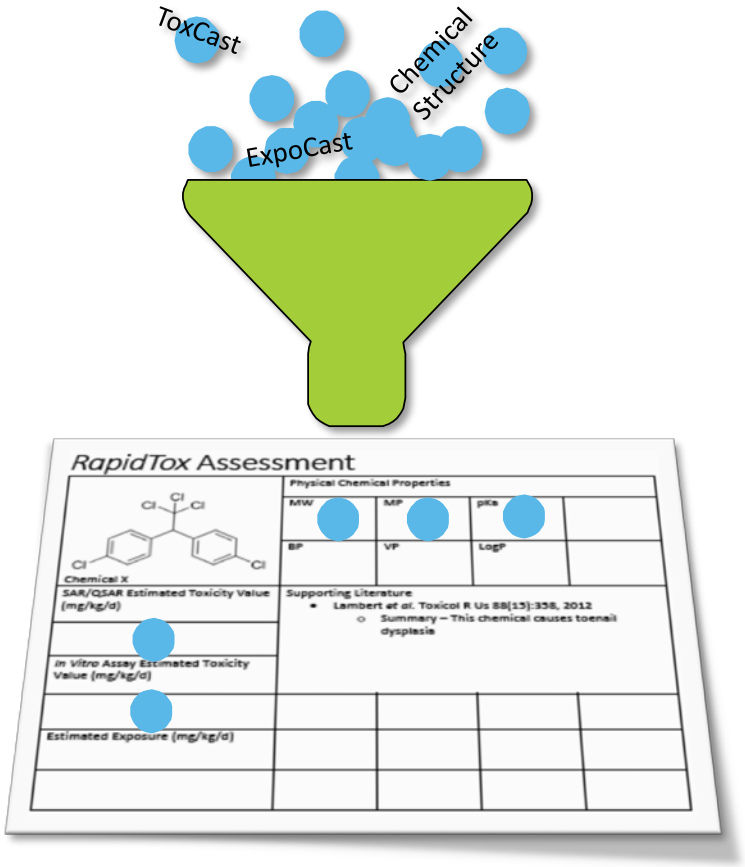
Future of Tox Testing Will Require Integrating HTS with Other Data



Application to Regulatory Decisions



Prioritization and Screening in EDSP



Development of Lower-Tier Assessment Products for Data Poor Chemicals

Acknowledgements

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

ORD Colleagues:
- NERL
- NHEERL
- NCEA

EPA's National Center for Computational Toxicology