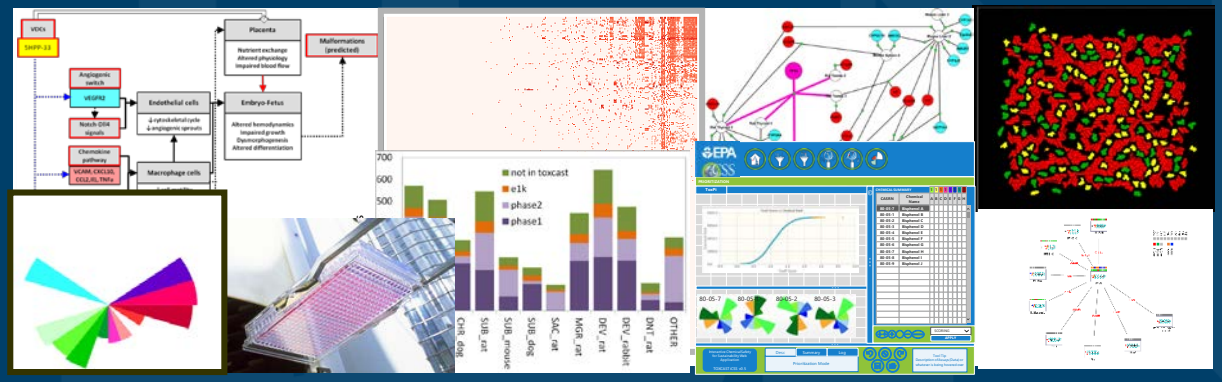
EPA United States Environmental Protection Agency

Building Scientific Confidence in the Development and Evaluation of Read-Across Using Tox21 Approaches



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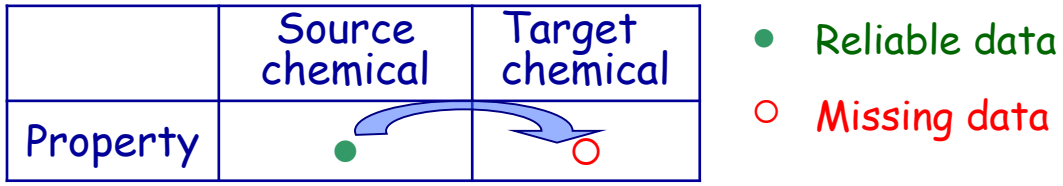
The views expressed in this presentation are those of the author and do not necessarily reflect the views or policies of the U.S. EPA

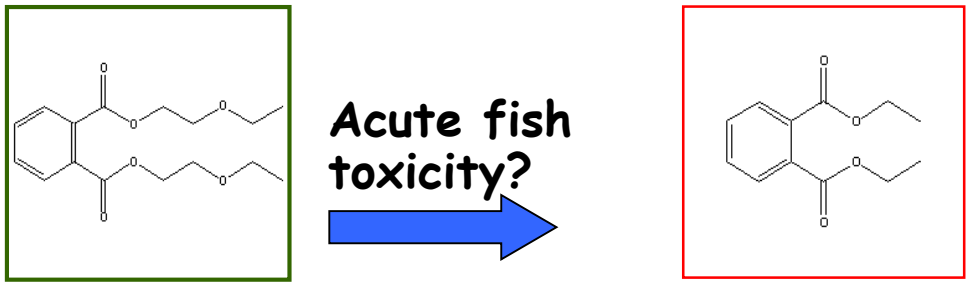
Outline

- Definitions
- Workflow for category development and read-across
- Identifying the sources of uncertainties associated with read-across and practical strategies to address these
- Quantifying uncertainties and Assessing Performance of read-across
- From research to implementation
- Summary

Definitions: Read-across

Known information on the property of a substance (source chemical) is used to make a prediction of the same property for another substance (target chemical) that is considered "similar" i.e. Endpoint & often study specific





Known to be harmful

Predicted to be harmful

Chemical category and read-across: General Workflow

1. Decision context
2. Data gap analysis
3. Overarching hypothesis
4. Analogue identification
5. Analogue evaluation
   - Data gap filling
6. Uncertainty assessment

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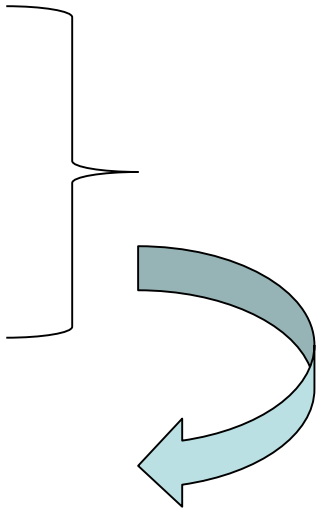
1. Decision context

- Prioritisation e.g. PMN
- Screening level hazard assessment
- Risk Assessment e.g PPRTV



- Different decision contexts will dictate the level of uncertainty that can be tolerated

6. Sources of Uncertainty

- Analogue or category approach? (#analogues)
- Data quality
- Overarching hypothesis/Similarity rationale – how to identify similar analogues and justify their similarity for the endpoint of interest
- Address the dissimilarities and whether these are significant from a toxicological standpoint
- Presence vs absence of toxicity
- Toxicokinetics – including Metabolism

Identifying Uncertainties

- Several pub
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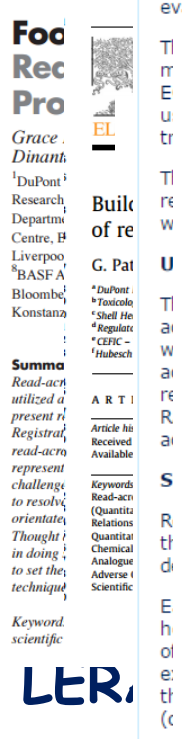
    - Frame
      (ECH
      2014, LER


Experts Work

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stakeholders.

Agenda for 3 Oct

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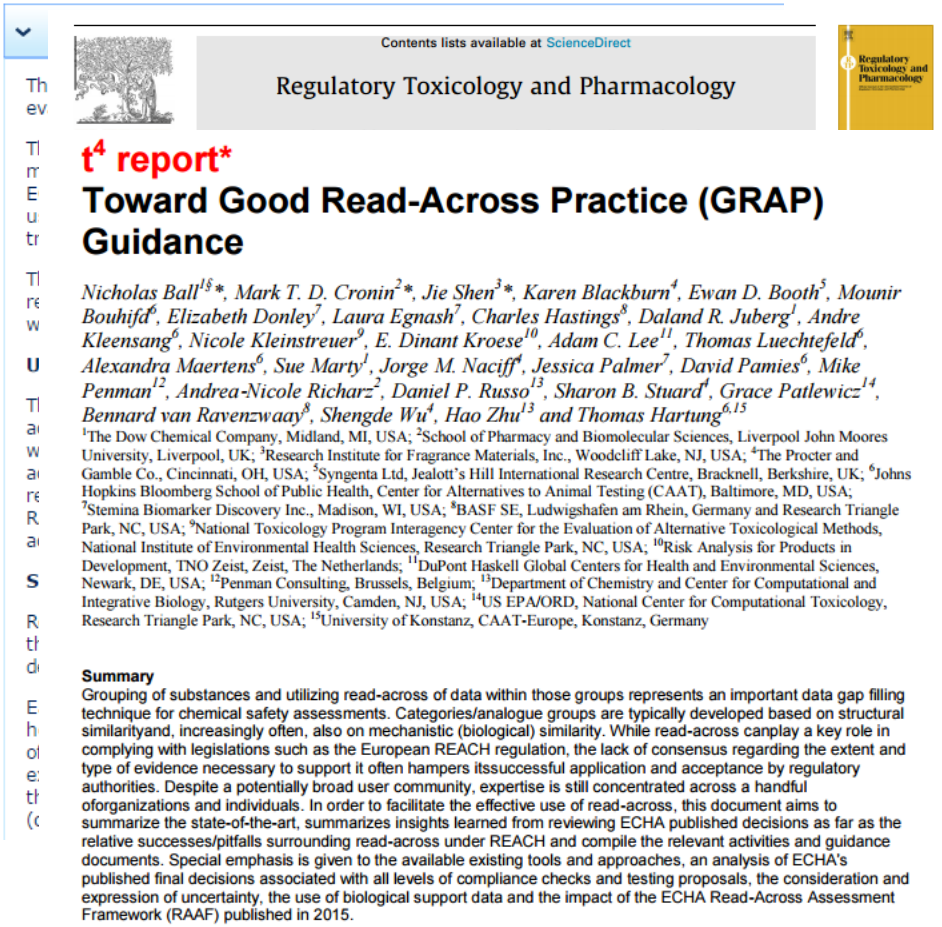
Contents lists available at ScienceDirect

Regulatory Toxicology and Pharmacology

t⁴ report*

Toward Good Read-Across Practice (GRAP) Guidance

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Summary

Grouping of substances and utilizing read-across of data within those groups represents an important data gap filling technique for chemical safety assessments. Categories/analogue groups are typically developed based on structural similarityand, increasingly often, also on mechanistic (biological) similarity. While read-across canplay a key role in complying with legislations such as the European REACH regulation, the lack of consensus regarding the extent and type of evidence necessary to support it often hampers itssuccessful application and acceptance by regulatory authorities. Despite a potentially broad user community, expertise is still concentrated across a handful oforganizations and individuals. In order to facilitate the effective use of read-across, this document aims to summarize the state-of-the-art, summarizes insights learned from reviewing ECHA published decisions as far as the relative successes/pitfalls surrounding read-across under REACH and compile the relevant activities and guidance documents. Special emphasis is given to the available existing tools and approaches, an analysis of ECHA's published final decisions associated with all levels of compliance checks and testing proposals, the consideration and expression of uncertainty, the use of biological support data and the impact of the ECHA Read-Across Assessment Framework (RAAF) published in 2015.

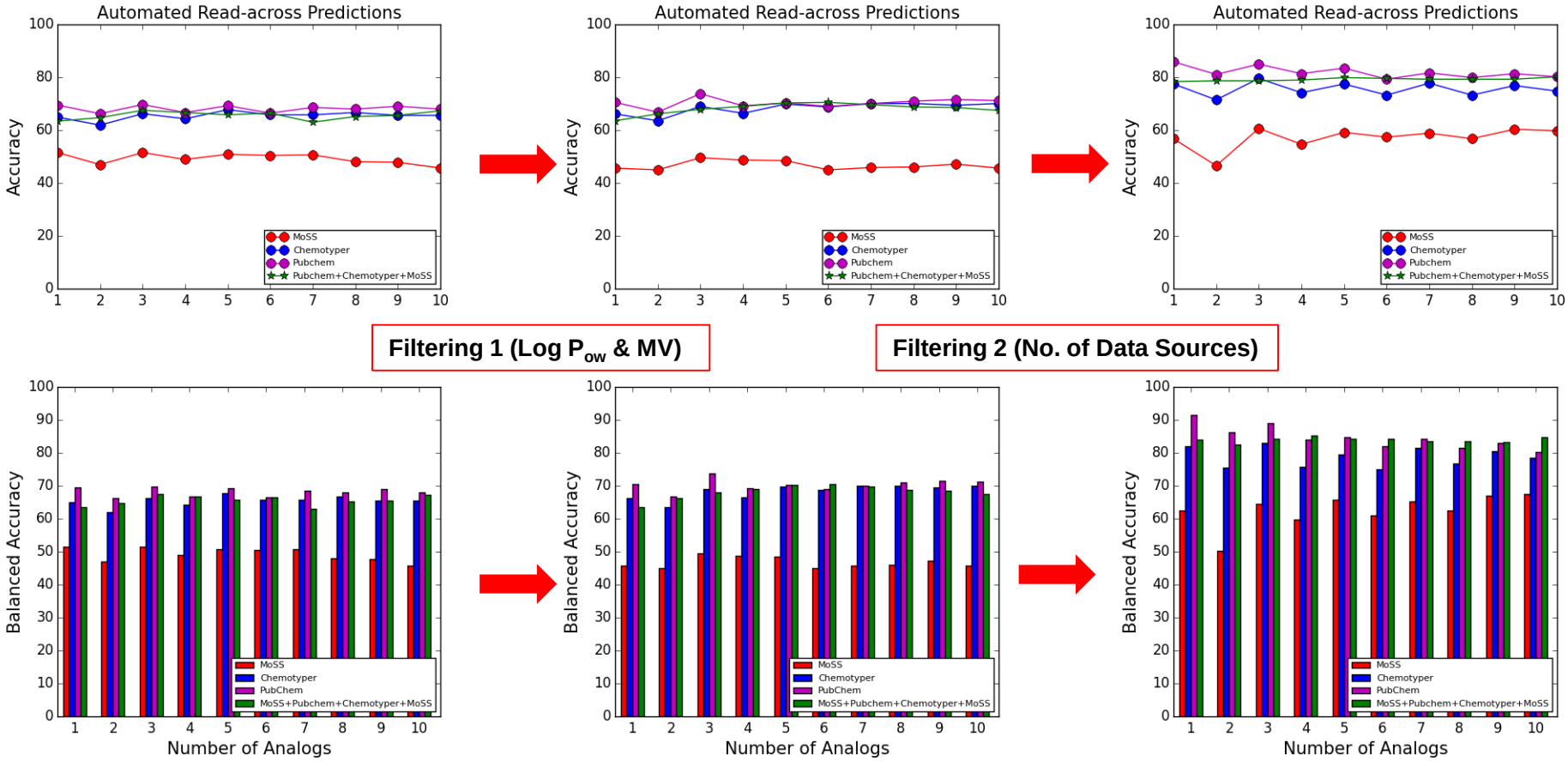
Addressing uncertainties - 1

- Search and Selection of analogues
- Using metabolism information
- Presence or absence of toxicity
- Using in vitro data such as HTS data to enhance read-across

Search and selection of analogues

- Explored the use of different structure-based approaches (Pubchem, Chemotyper and MoSS MCSS with Tanimoto index as a measure of similarity) to identify hindered phenol analogues and evaluate their validity for reading across Estrogenicity
- Make a read-across Estrogenicity prediction for each target hindered phenol

Read-across predictions



Slide from P Pradeep

Case study conclusions

- Initial selection of analogues based on different descriptor sets (for this example) was invariant to the read-across prediction performance

- Evaluating analogue validity paying close attention to the quality of the underlying analogue data and relevant physchem properties did significantly improve read-across predictive performance

Low toxicity

- Case studies focused on HPV categories
  - Long chain alcohols (LCAs)
  - Ethylene glycols
- Mixed outcomes relative to the adverse outcomes that proved to be most sensitive in driving the risk assessment
- If mechanism is known then HTS data from ToxCast appeared to substantiate the read-across (i.e., irritation)
- HTS data of less value when metabolites are implicated (e.g., ethylene glycols and associated renal and repro effects)

Addressing uncertainities - 2

- Read-across acceptance is context dependent – based on subjective expert judgement assessment – potential lack of harmonised or reproducible decisions
- No clear understanding of what constitutes success
- Do we know what the performance of a read-across is really like on a more general level?

Critical need is an objective measure of uncertainty in a read-across prediction

Quantifying uncertainty & Assessing performance of read-across

- GenRA (Generalised Read-Across) is a "local validity" approach
- Predicting toxicity as a similarity-weighted activity of nearest neighbours based on chemistry and bioactivity descriptors

$$y_i^{tox} = \frac{\sum_j^k s_{ij}^{\alpha} x_j^{tox}}{\sum_j^k s_{ij}^{\alpha}}$$

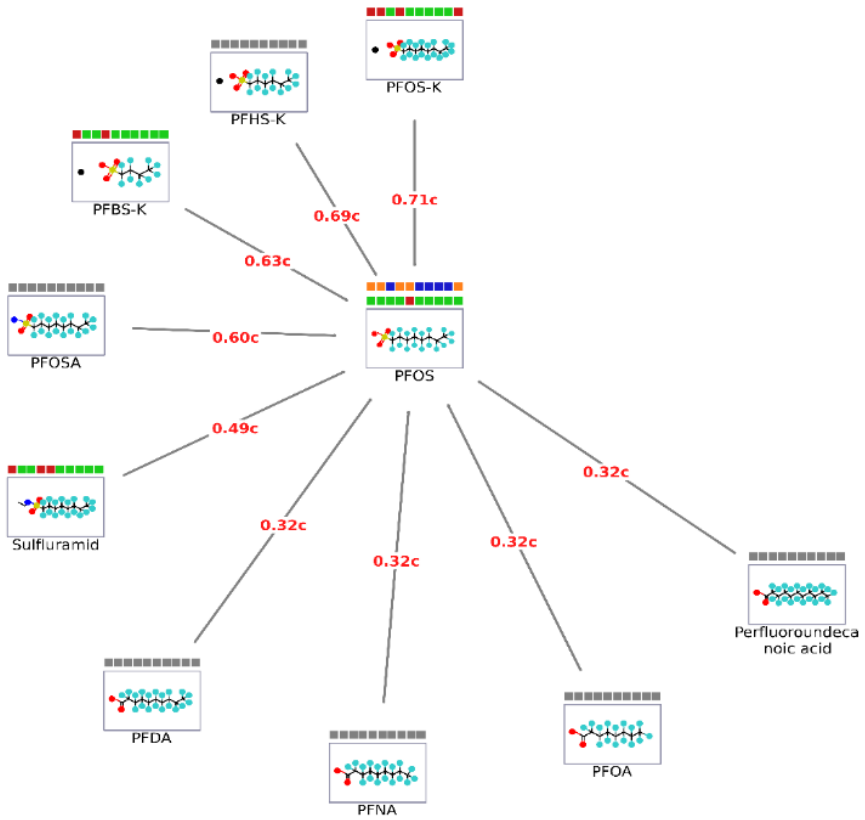
$$\alpha = \{chm, bio, bc\}$$

Where x_j^{tox}, in this case, is the *in vivo* toxicity of chemical *j*

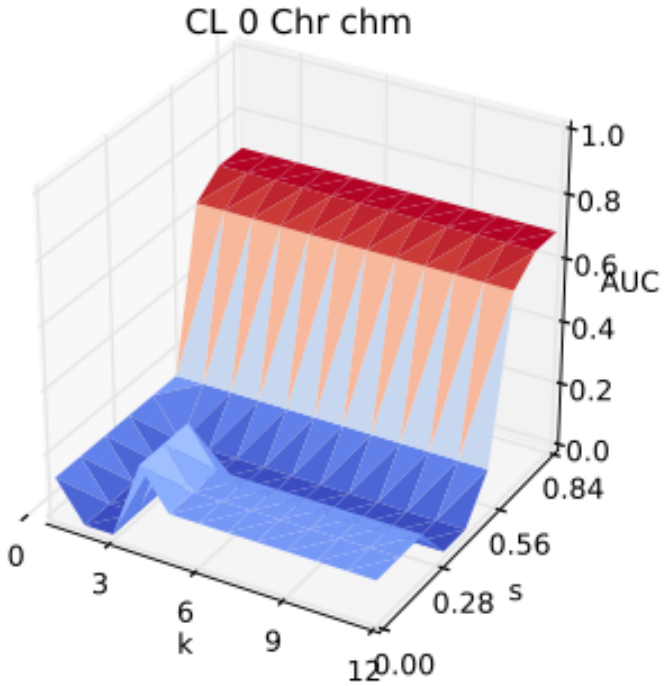
Shah et al, submitted

- Initial focus relied on standard guideline studies
- Endpoint recorded as binary outcomes

GenRA: Nominal cluster



Explore performance as a function of number of nearest neighbours or similarity index



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Quantifying uncertainty & Assessing performance of read-across

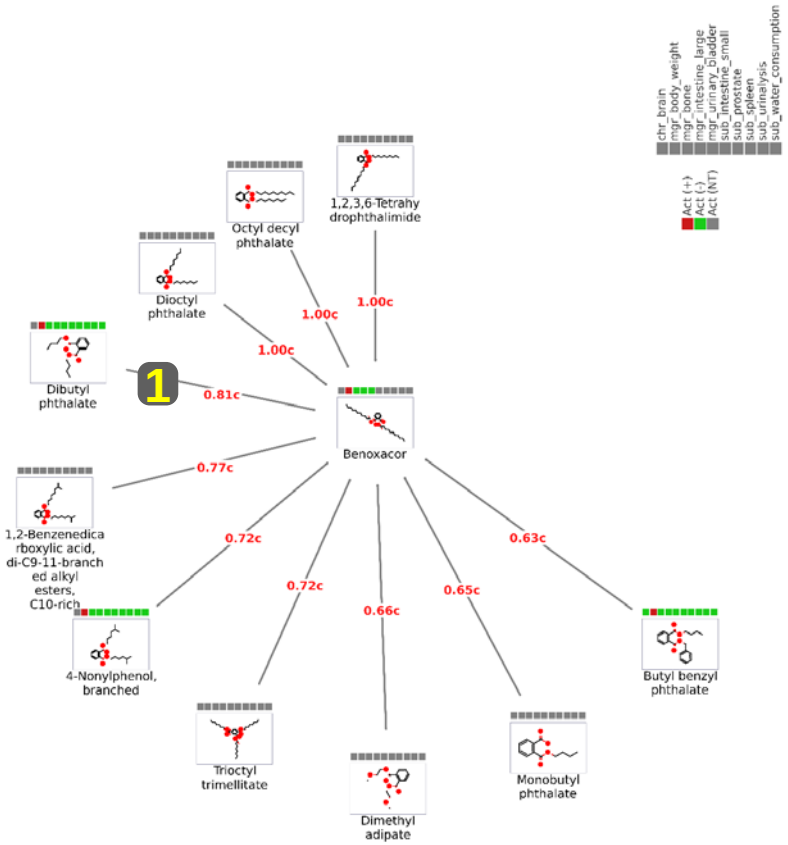
- Tested and compared
  1. Chemical descriptors
  2. Bioactivity descriptors
  3. Hybrid of chemical and bioactivity descriptors
- No preselection of descriptors was performed
- Bioactivity descriptors were often found to be more predictive of in vivo toxicity outcomes
- The approach enabled a performance baseline for read-across predictions of specific study outcomes to be established
- But still context dependent on the endpoint and the chemical neighbourhood under study

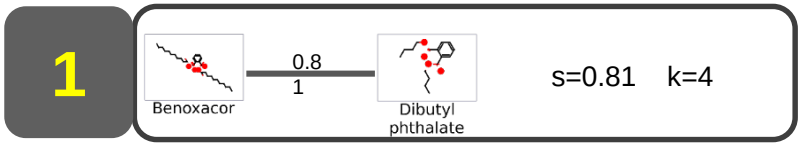
Quantifying uncertainty & Assessing performance of read-across

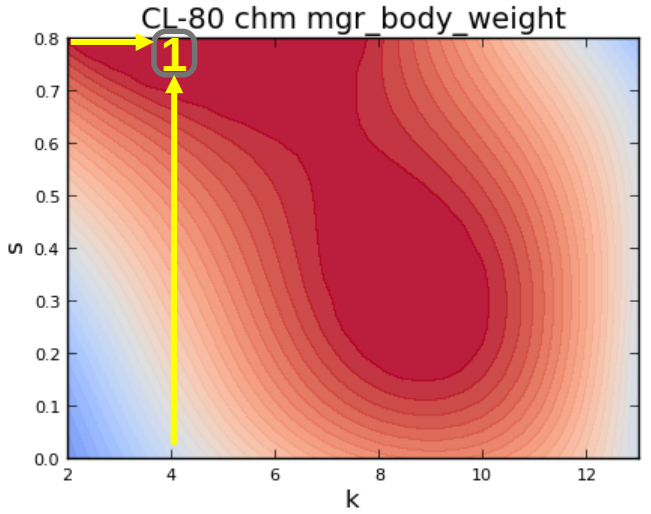
- Next steps in progress:

- Predicting endpoint outcomes at a target organ level rather than as binary summary outcomes and applying the approach in practice
- Use of other chemical descriptor sets that encode more expert knowledge of SARs
- Incorporating TK information

Analysing local neighborhood of a chemical







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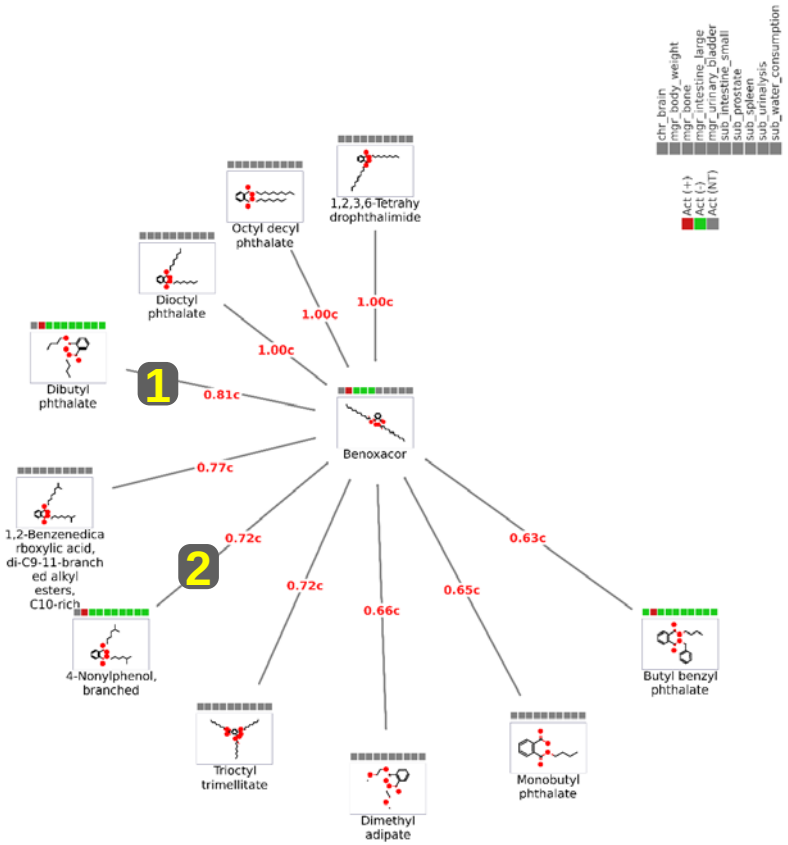

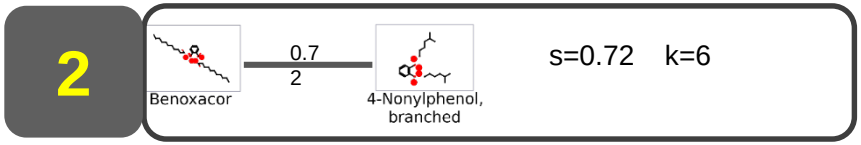

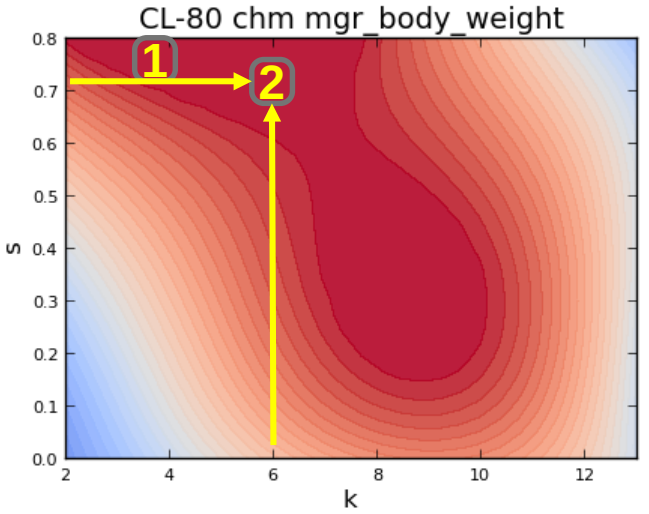

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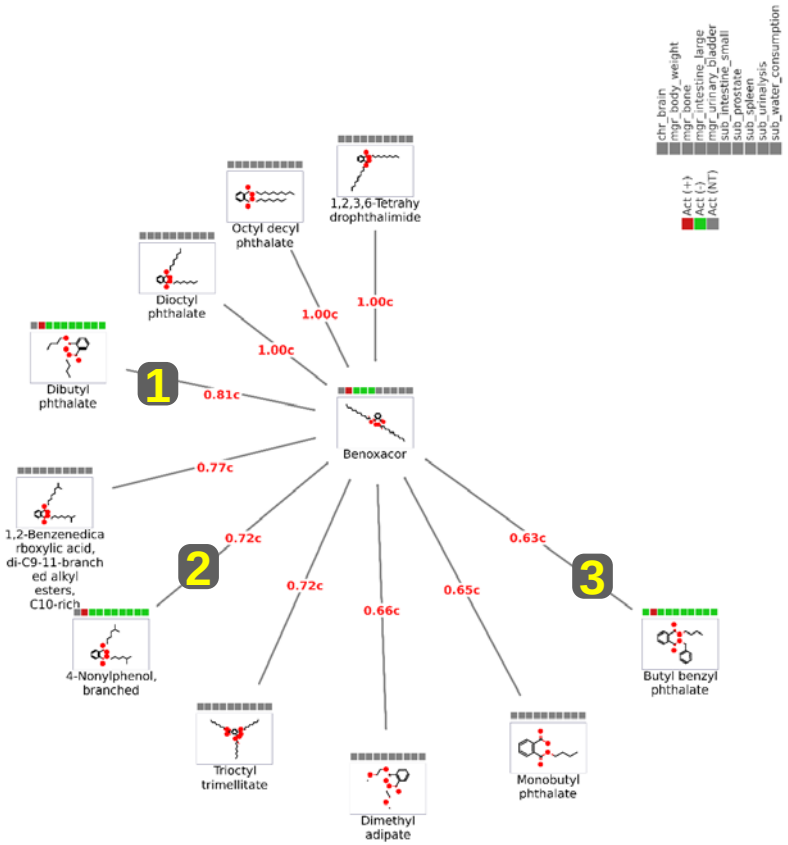

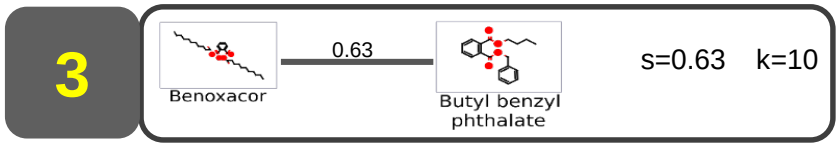

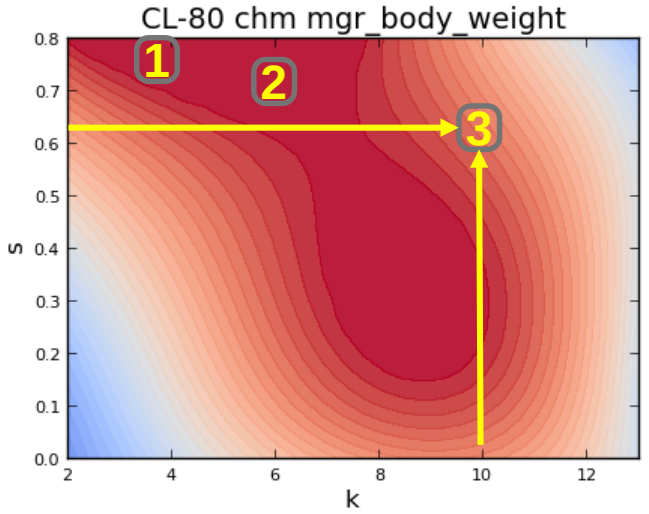

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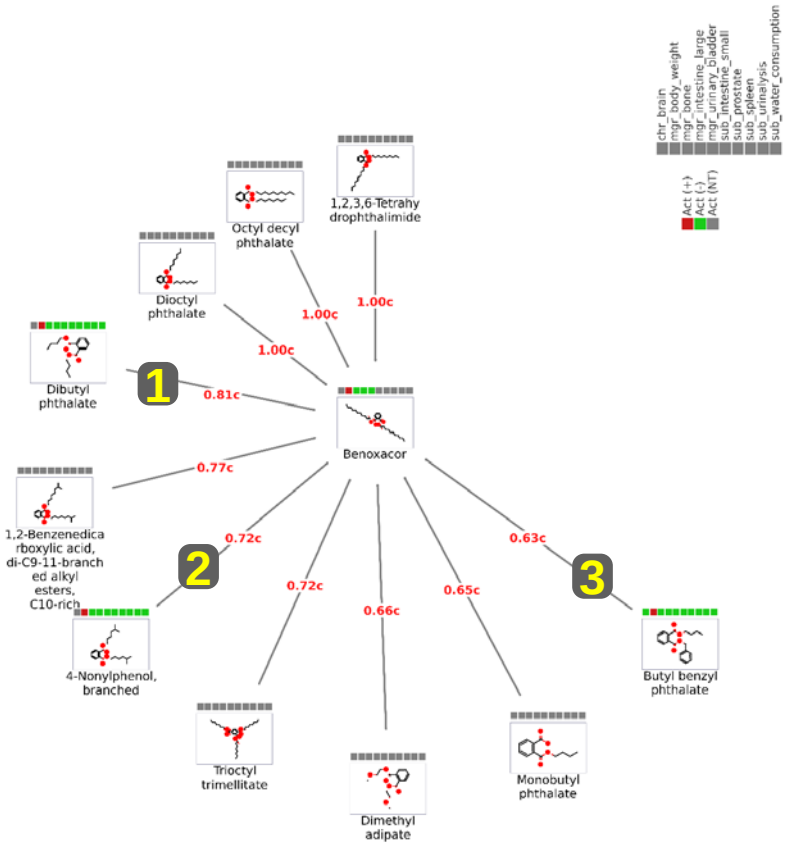
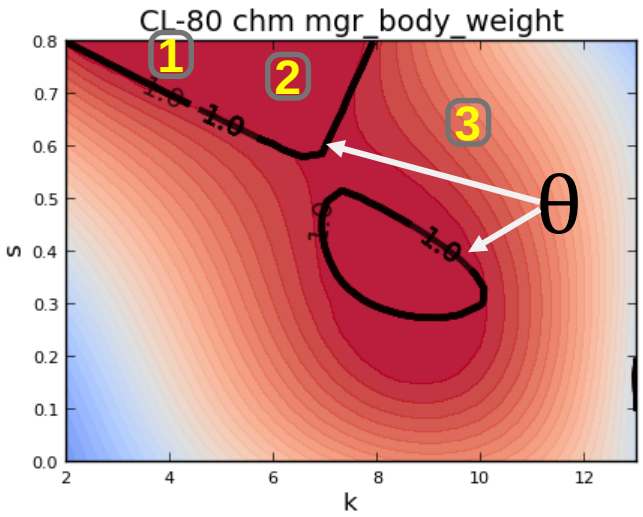
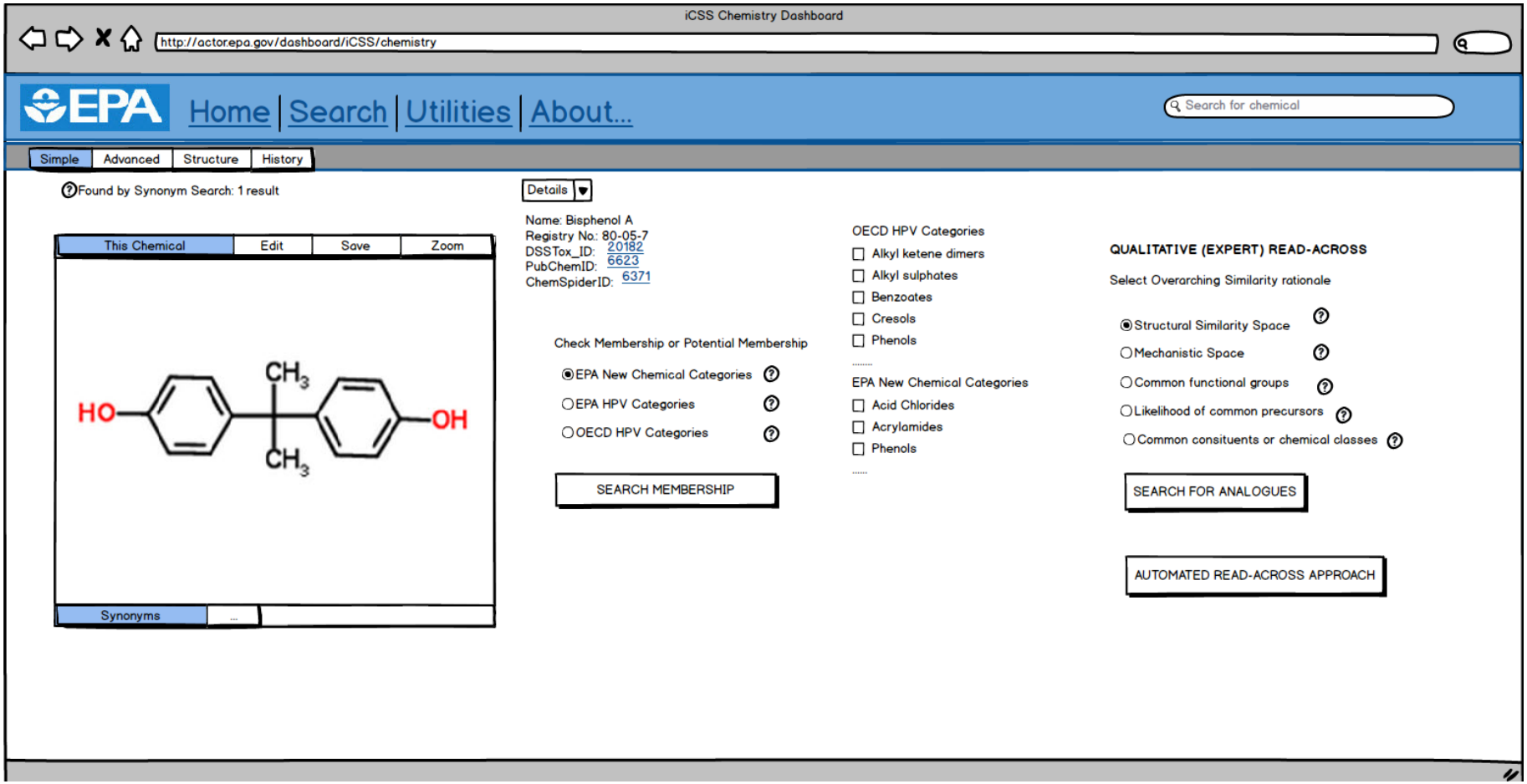




From Research to Implementation

- Public accessible tool building on the iCSS Chemistry Dashboard under development



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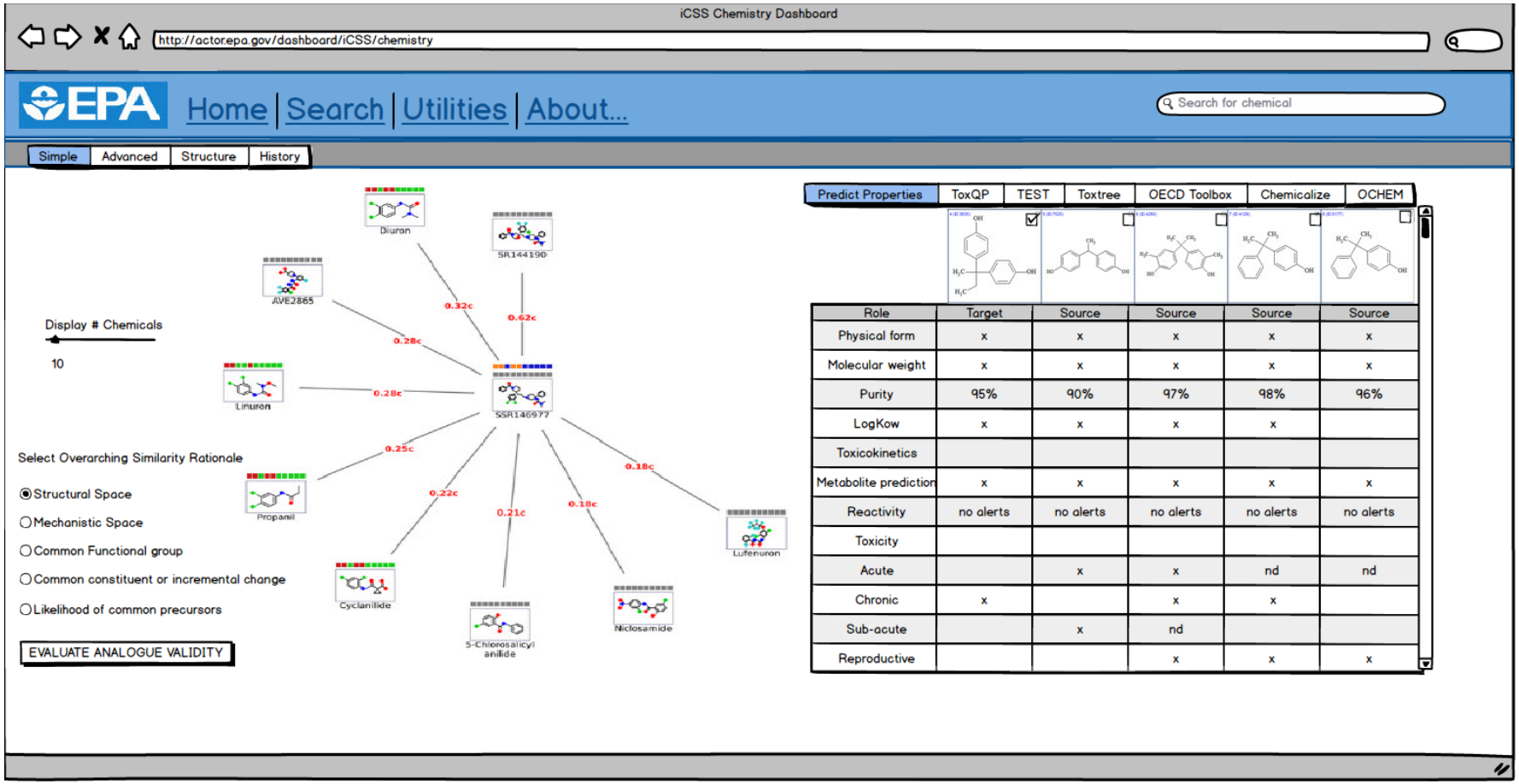

Role	Target	Source	Source	Source	Source
Physical form	x	x	x	x	x
Molecular weight	x	x	x	x	x
Purity	95%	90%	97%	98%	96%
LogKow	x	x	x	x	
Toxicokinetics					
Metabolite prediction	x	x	x	x	x
Reactivity	no alerts	no alerts	no alerts	no alerts	no alerts
Toxicity					
Acute		x	x	nd	nd
Chronic	x		x	x	
Sub-acute		x	nd		
Reproductive			x	x	x

EPA United States Environmental Protection Agency

Summary

- Still many challenges remain in read-across
- Quantifying the uncertainty of read-across prediction is a critical issue
- Have illustrated a handful of the research directions being taken

Acknowledgements

- Imran Shah
- Tony Williams
- Richard Judson
- Rusty Thomas