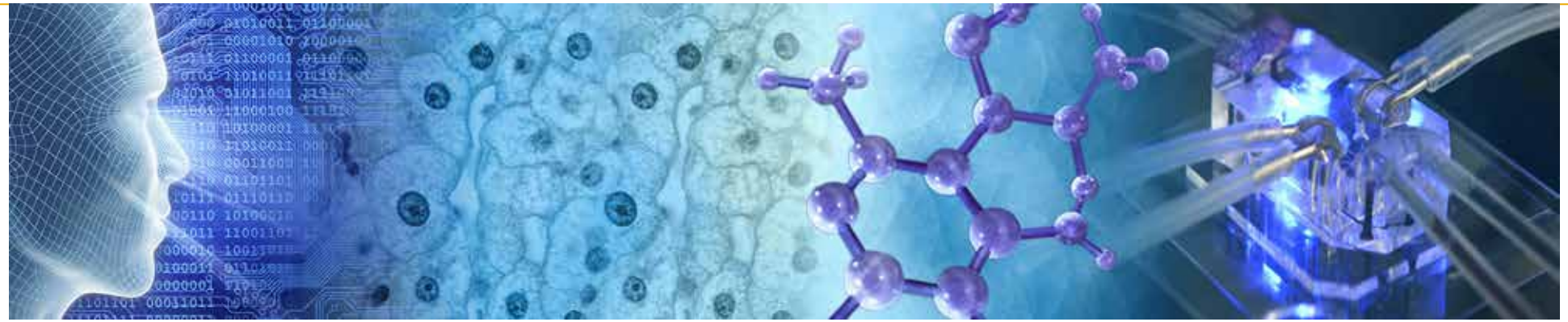
NTP
National Toxicology Program



International Collaboration to Build Predictive Models for Acute Oral Toxicity

Kamel Mansouri
LEAD COMPUTATIONAL CHEMIST, ILS IN SUPPORT OF NICEATM

SOT 2019 – Abstract: 2563

Disclaimer: ILS staff provide technical support for NICEATM, but do not represent NIEHS, NTP, or the official positions of any federal agency. (the author declares no conflict of interest)

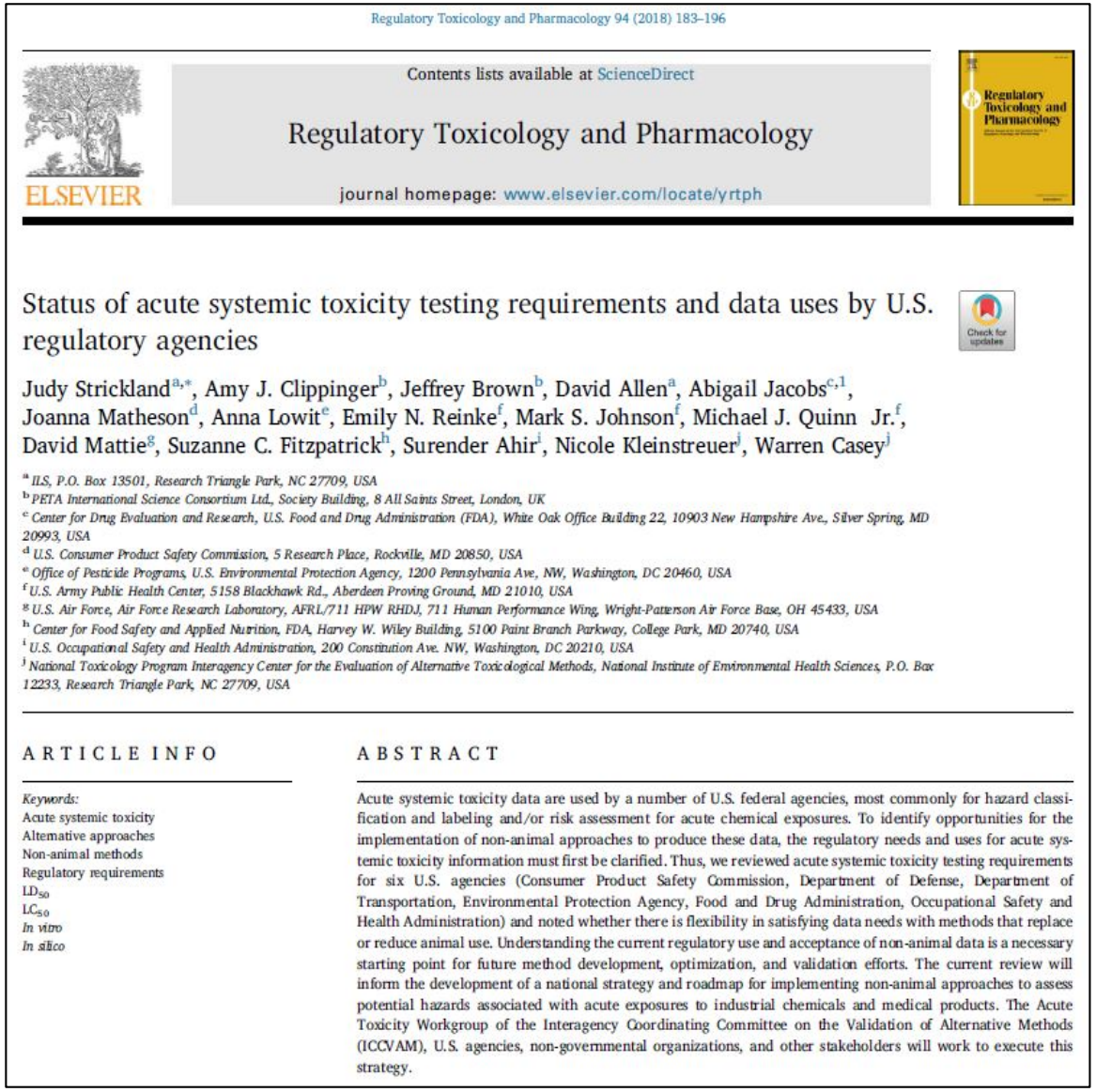
Overview

- Project scope: acute oral toxicity
  - Regulatory use of these data
  - Endpoints selected for predictive modeling
  - Compiling inventory of rat acute oral LD50
  - Establishing training, evaluation, and prediction sets
  - Evaluation of submitted models
- International contributors
- Generation of consensus predictions
- Current status and public release

Scoping Regulatory Needs

ICCVAM Acute Toxicity Workgroup

- Identifies federal agency requirements, needs, and decision contexts for using acute systemic toxicity data



Regulatory Toxicology and Pharmacology 94 (2018) 183–196

Contents lists available at ScienceDirect

Regulatory Toxicology and Pharmacology

journal homepage: www.elsevier.com/locate/yrtph

Status of acute systemic toxicity testing requirements and data uses by U.S. regulatory agencies

Judy Strickland[a,*], Amy J. Clippinger[b], Jeffrey Brown[b], David Allen[a], Abigail Jacobs[c,1], Joanna Matheson[d], Anna Lowit[e], Emily N. Reinke[f], Mark S. Johnson[f], Michael J. Quinn Jr.[f], David Mattie[g], Suzanne C. Fitzpatrick[h], Surender Ahir[i], Nicole Kleinstreuer[j], Warren Casey[j]

[a] ILS, P.O. Box 13501, Research Triangle Park, NC 27709, USA
[b] PETA International Science Consortium Ltd., Society Building, 8 All Saints Street, London, UK
[c] Center for Drug Evaluation and Research, U.S. Food and Drug Administration (FDA), White Oak Office Building 22, 10903 New Hampshire Ave., Silver Spring, MD 20993, USA
[d] U.S. Consumer Product Safety Commission, 5 Research Place, Rockville, MD 20850, USA
[e] Office of Pesticide Programs, U.S. Environmental Protection Agency, 1200 Pennsylvania Ave., NW, Washington, DC 20460, USA
[f] U.S. Army Public Health Center, 5158 Blackhawk Rd., Aberdeen Proving Ground, MD 21010, USA
[g] U.S. Air Force, Air Force Research Laboratory, AFRL/711 HPW RHDJ, 711 Human Performance Wing, Wright-Patterson Air Force Base, OH 45433, USA
[h] Center for Food Safety and Applied Nutrition, FDA, Harvey W. Wiley Building, 5100 Paint Branch Parkway, College Park, MD 20740, USA
[i] U.S. Occupational Safety and Health Administration, 200 Constitution Ave. NW, Washington, DC 20210, USA
[j] National Toxicology Program Interagency Center for the Evaluation of Alternative Toxicological Methods, National Institute of Environmental Health Sciences, P.O. Box 12233, Research Triangle Park, NC 27709, USA

ARTICLE INFO

Keywords:
Acute systemic toxicity
Alternative approaches
Non-animal methods
Regulatory requirements
LD_{50}
LC_{50}
In vitro
In silico

ABSTRACT

Acute systemic toxicity data are used by a number of U.S. federal agencies, most commonly for hazard classification and labeling and/or risk assessment for acute chemical exposures. To identify opportunities for the implementation of non-animal approaches to produce these data, the regulatory needs and uses for acute systemic toxicity information must first be clarified. Thus, we reviewed acute systemic toxicity testing requirements for six U.S. agencies (Consumer Product Safety Commission, Department of Defense, Department of Transportation, Environmental Protection Agency, Food and Drug Administration, Occupational Safety and Health Administration) and noted whether there is flexibility in satisfying data needs with methods that replace or reduce animal use. Understanding the current regulatory use and acceptance of non-animal data is a necessary starting point for future method development, optimization, and validation efforts. The current review will inform the development of a national strategy and roadmap for implementing non-animal approaches to assess potential hazards associated with acute exposures to industrial chemicals and medical products. The Acute Toxicity Workgroup of the Interagency Coordinating Committee on the Validation of Alternative Methods (ICCVAM), U.S. agencies, non-governmental organizations, and other stakeholders will work to execute this strategy.

Agency-Based Modeling Endpoint Selection

Binary Models

Hazard

- Highly toxic (≤50 mg/kg)
- Toxic (>50-5000 mg/kg)

+ Nontoxic (>2000 mg/kg)

Continuous Model

Point estimates of LD50 values

Categorical Models

EPA Categories

Hazard

- I (≤ 50 mg/kg)
- II (>50 ≤ 500 mg/kg)
- III (>500 ≤ 5000 mg/kg)
- IV (>5000 mg/kg)

GHS Categories

Packing Group

- I (≤ 5 mg/kg)
- II (>5 ≤ 50 mg/kg)
- III (>50 ≤ 300 mg/kg)
- IV (>300 ≤ 2000 mg/kg)
- NC (> 2000 mg/kg)

OSHA

Hazard

Available data for modeling

Rat oral LD50s:
16,297 chemicals total
34,508 LD50 values

15,688 chemicals total
21,200 LD50 values

QSAR-ready standardization

Desalted, stereochemistry stripped, tautomers and nitro groups standardized, valence corrected, structures neutralized

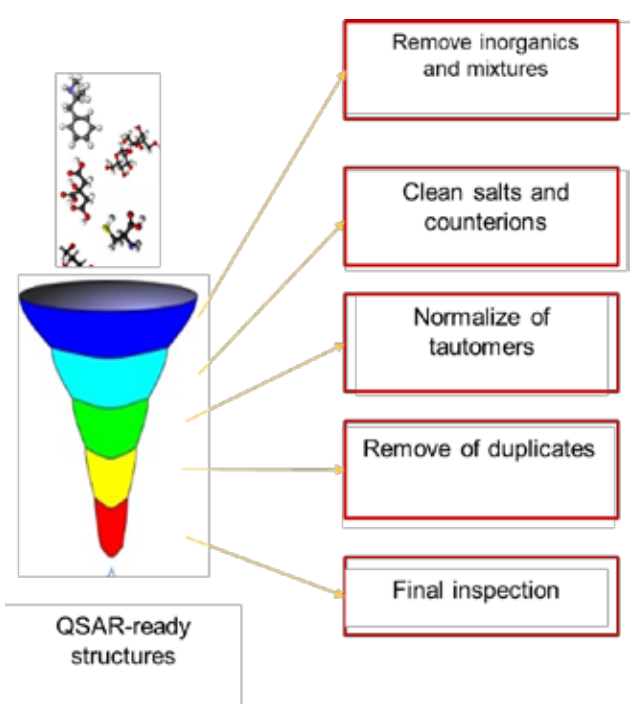
11992 chemicals with accurate structures

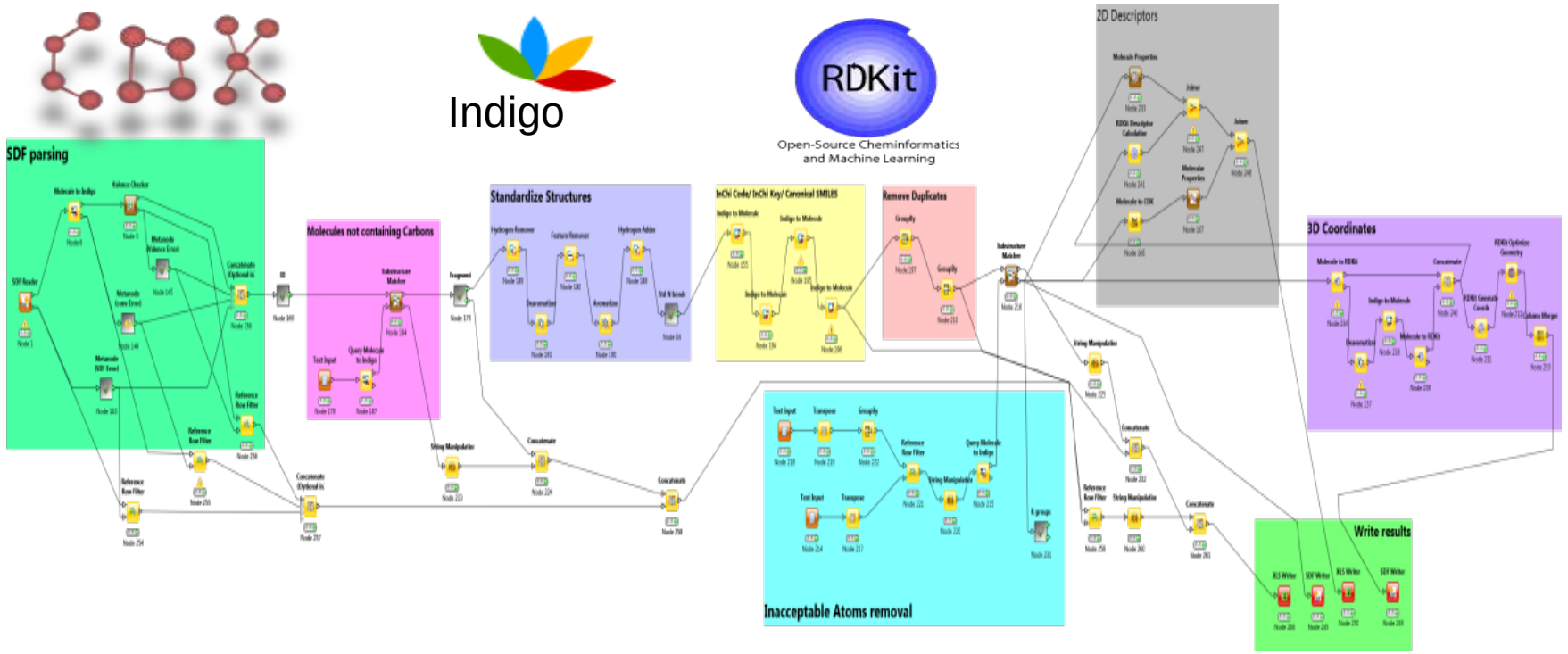
- Very toxic endpoint: 11886 entries (binary, 0/1)
- Non-toxic endpoint: 11871 entries (binary, 0/1)
- EPA endpoint: 11755 entries (categorical, 4 categories)
- GHS endpoint: 11845 entries (categorical, 5 categories)
- LD50 endpoint: 8908 entries (continuous values)

QSAR-ready KNIME workflow

Aim of the workflow:

- Combine different procedures and ideas
- Minimize the differences between the structures used for prediction
- Produce a flexible free and open source workflow to be shared





Fourches et al. J Chem Inf Model, 2010, 29, 476 – 488

Wedebye et al. Danish EPA Environmental Project No. 1503, 2013

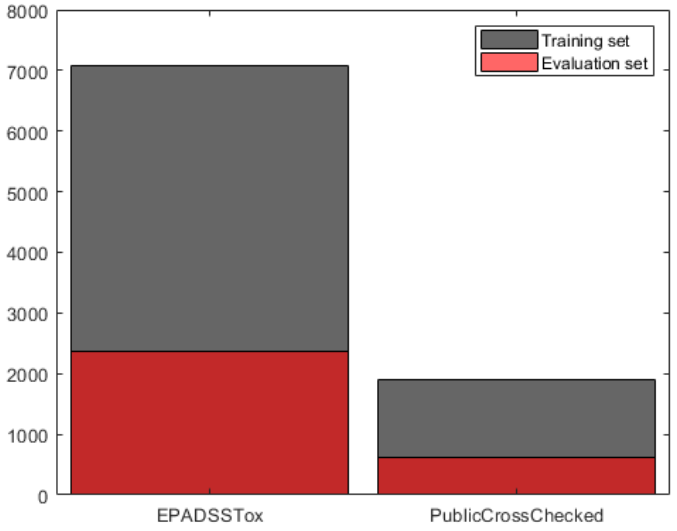
Mansouri et al. (http://ehp.niehs.nih.gov/15-10267/)

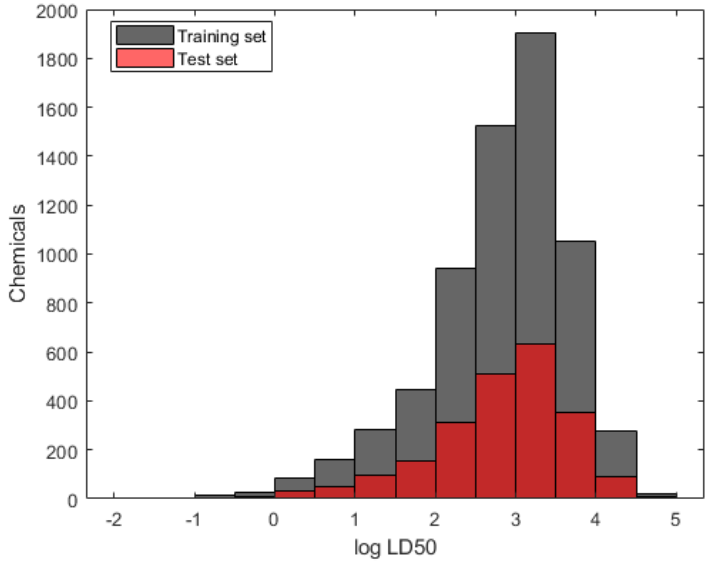
Establishing Modeling Dataset

- **Training and evaluation sets:**

- 11,992 chemicals from the final inventory of chemicals with QSAR-ready structures having rat oral acute toxicity data were split into training and test sets:
  - 75% training set: 8,994 chemicals
  - 25% evaluation set: 2,998 chemicals

- The same training and test chemicals across all endpoints
- Similar distributions and variability for values and categories
- Similar distribution of chemical structures sources





Establishing Modeling Dataset

- **Prediction set:**

Included Lists:

- ToxCast/Tox21
- EDSP
- TSCA
- Substances on the market (EPA Dashboard list)

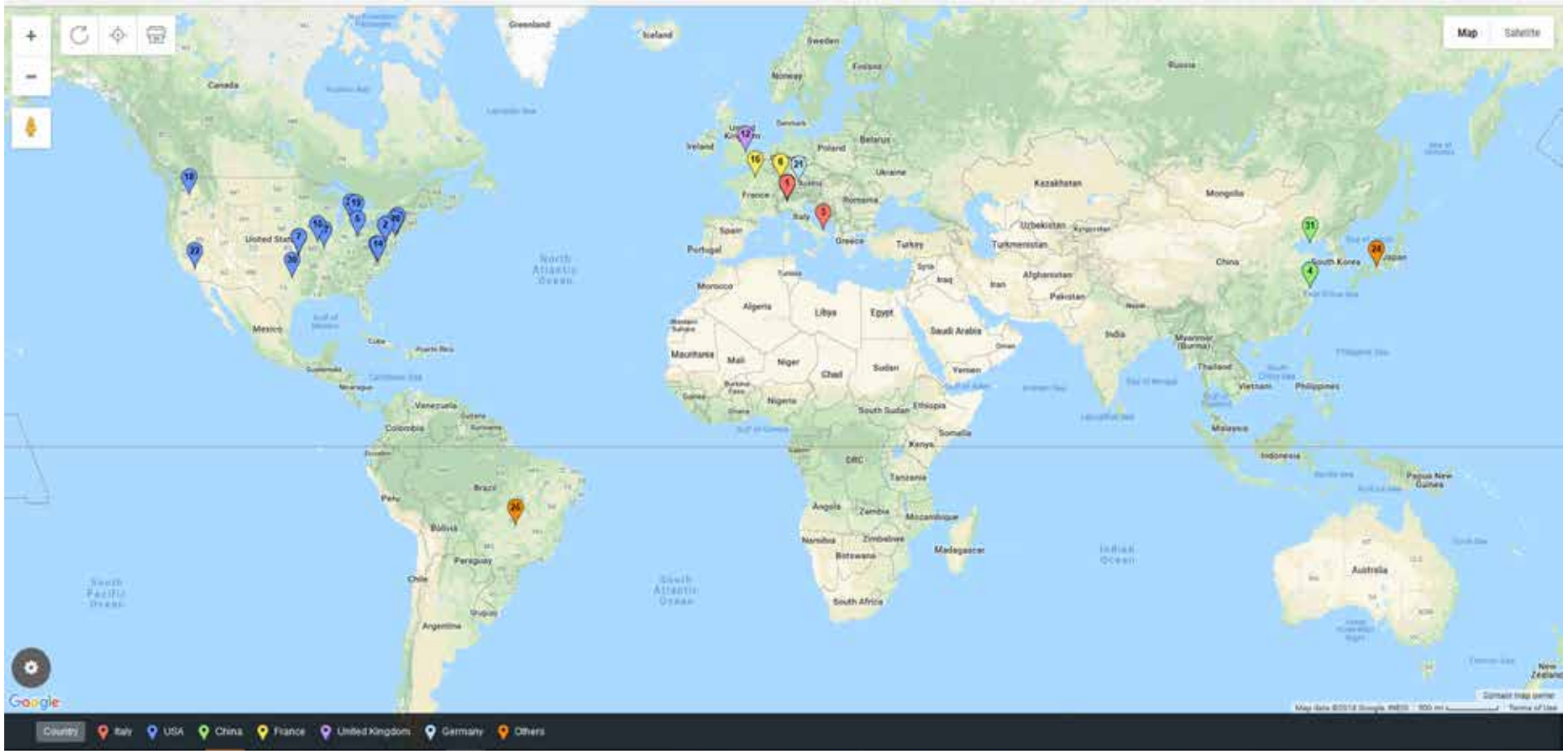
After QSAR-ready standardization:

48137 structures to be predicted (including the evaluation set)

International Collaboration

Consortium:

- Participants from around the globe representing academia, industry, and government contributed



(https://batchgeo.com/map/d06c5d497ed8f76ecfee500c2b0e1dfa)

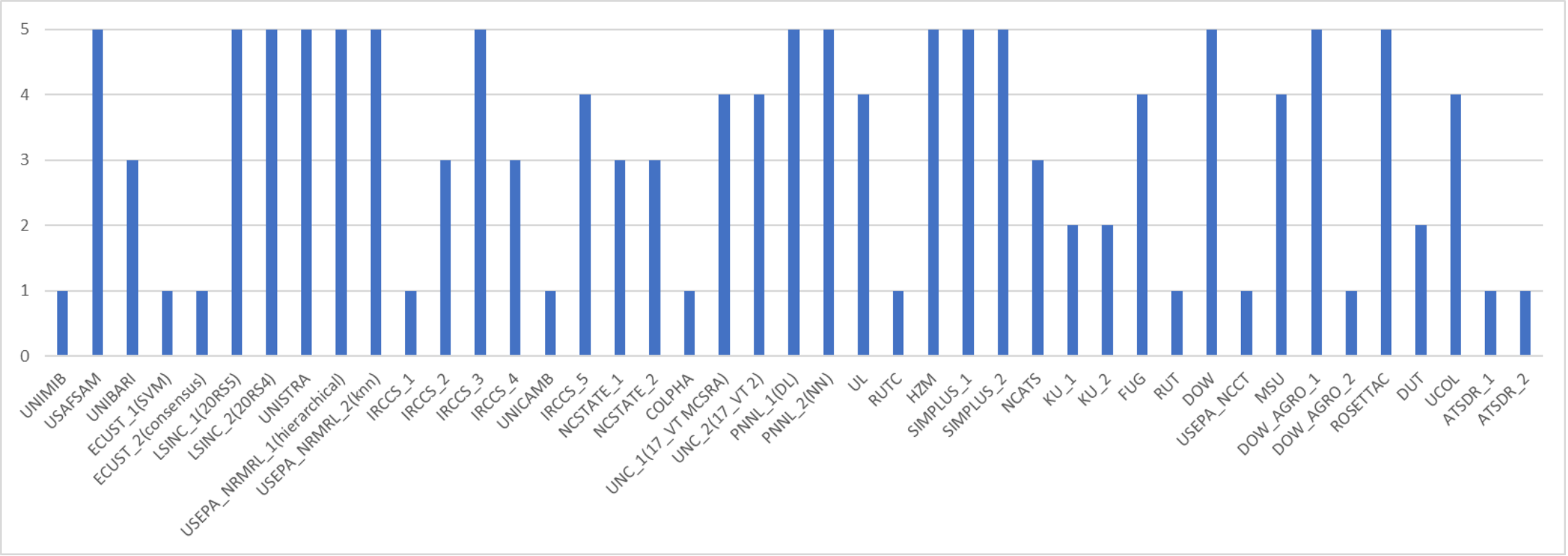
Submitted Models

- GHS categories: 23 models

Consortium Comprised 35 Participants/Groups

- Very Toxic: 32 models
- Non-toxic: 33 models
- EPA categories: 26 models
- LD50: 25 models

Total: 139 models



Evaluation procedure

Qualitative evaluation:

- Documentation
- Defined endpoint
- Unambiguous algorithm
- Availability of code
- Applicability domain definition
- Availability of data used for modeling
- Mechanistic interpretation

Quantitative evaluation:

- Goodness of fit: training statistics
- Evaluation set predictivity: statistics on the evaluation set
- Robustness: balance between (Goodness of fit) & (Test set predictivity)

$$S = 0.3 * (Goodness\ of\ fit) + 0.45 * (Test\ set\ predictivity) + 0.25 * (Robustness)$$

Categorical models (binary and multi-class):

$$Goodness\ of\ fit = 0.7 * (BA_{Tr}) + 0.3 * \left(1 - |\widehat{Sn_{Tr}} -$$

$$Test\ set\ predictivity = 0.7 * (BA_{Tst}) + 0.3 * \left(1 - |Sn_{Ts}\right.$$

$$Robustness = 1 - |BA_{Tr} - BA_{Tst}|$$

Continuous models:

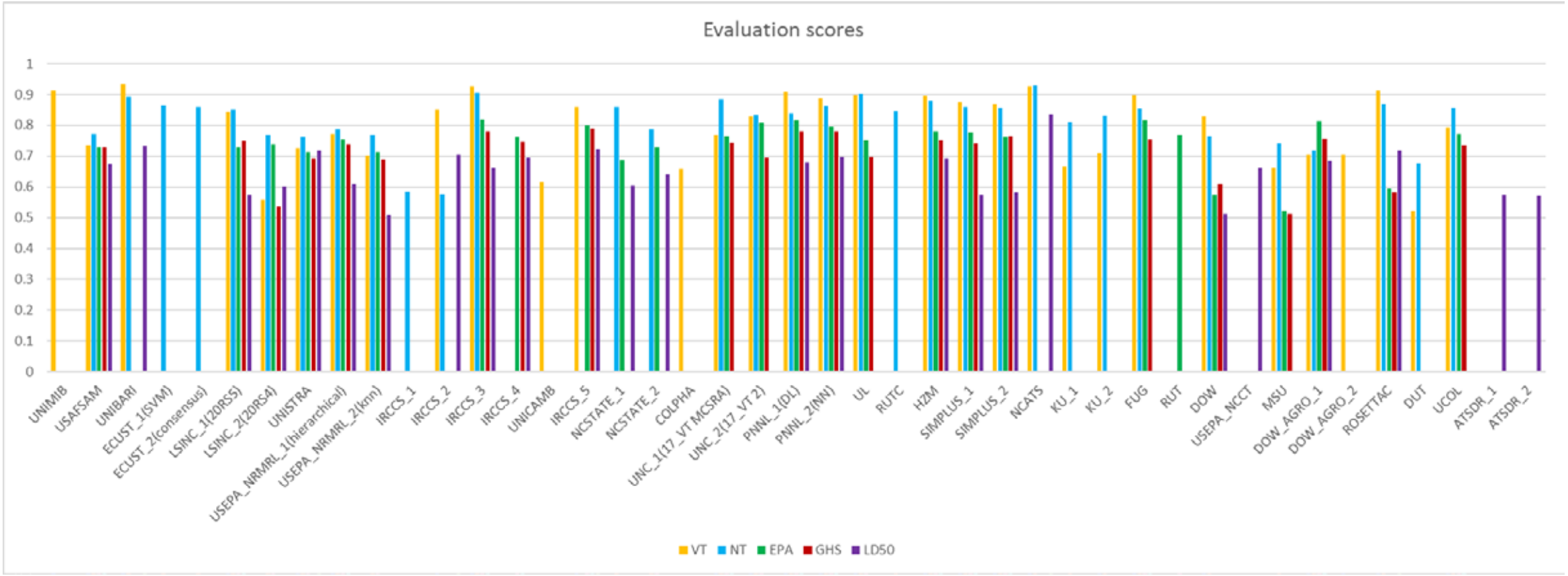
$$Goodness\ of\ fit = R^2_{Tr}$$

$$Test\ set\ predictivity = R^2_{Tst}$$

$$Robustness = 1 - |R^2_{Tr} - R^2_{Tst}|$$

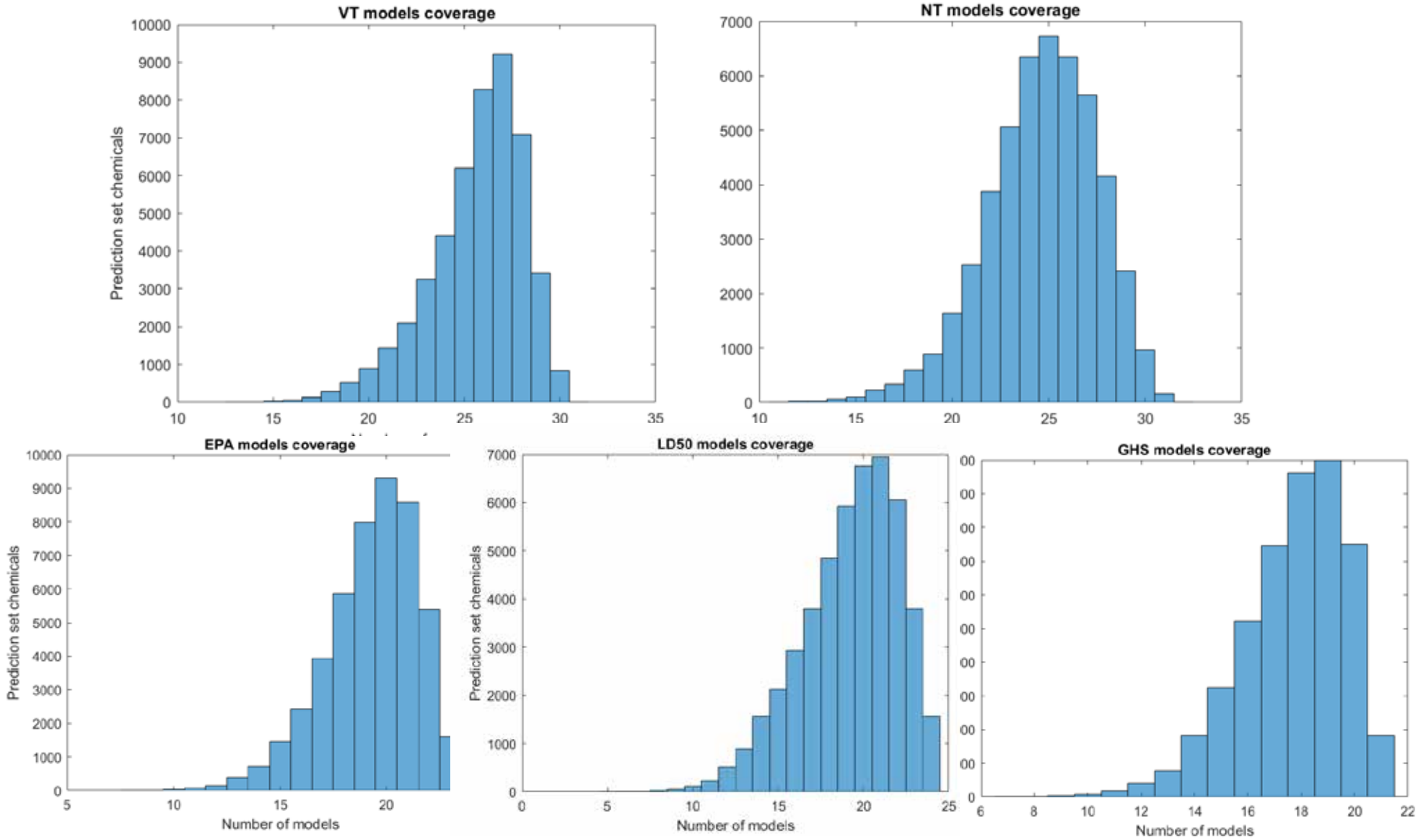
Evaluation results

Quantitative evaluation



Coverage of the models

Distribution of the number of models/chemical



Generation of Consensus Predictions

- Models passing qualitative evaluation (requirement for transparency; description of approach was sufficient)
- Integrating only *in-domain* predictions across chemicals in the prediction set (48,137 chemicals) for each model, respectively
  - Categorical models: weighted majority rule
  - Continuous model: weighted average

Kleinstreuer et al., Predictive models for acute oral systemic toxicity: A workshop to bridge the gap from research to regulation, Computational Toxicology, 2018, https://doi.org/10.1016/j.comtox.2018.08.002.

Performance Assessment

Consensus Model Statistics

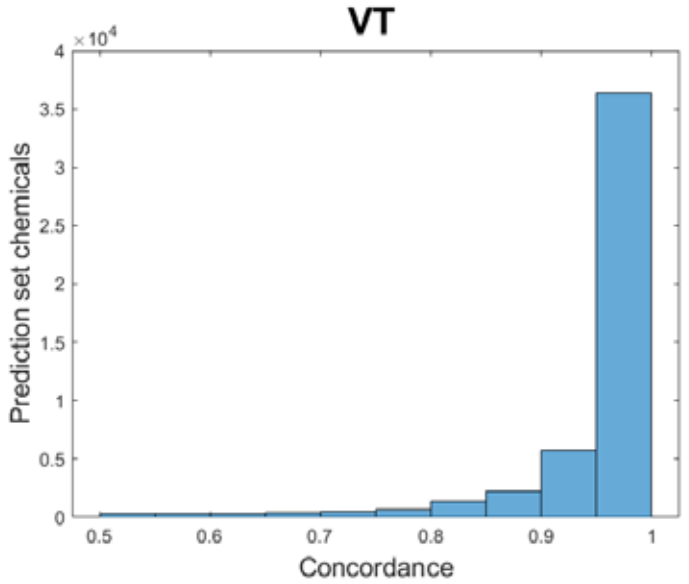
	Very Toxic		Non-Toxic		EPA		GHS	
	Train	Eval	Train	Eval	Train	Eval	Train	Eval
Sensitivity	0.87	0.67	0.93	0.70	0.73	0.50	0.63	0.45
Specificity	0.94	0.96	0.96	0.88	0.96	0.91	0.91	0.92
Balanced Accuracy	0.93	0.81	0.94	0.79	0.83	0.71	0.77	0.68
***In vivo* Balanced Accuracy**	0.81		0.89		0.82		0.79	

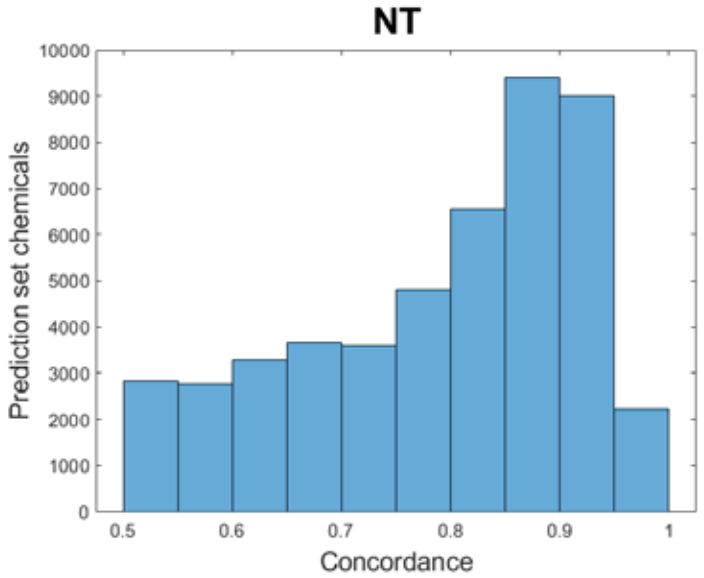
	LD50 values		LD50 values
	Train	Eval	***In Vivo***
R2	0.84	0.64	0.80
RMSE	0.32	0.51	0.42

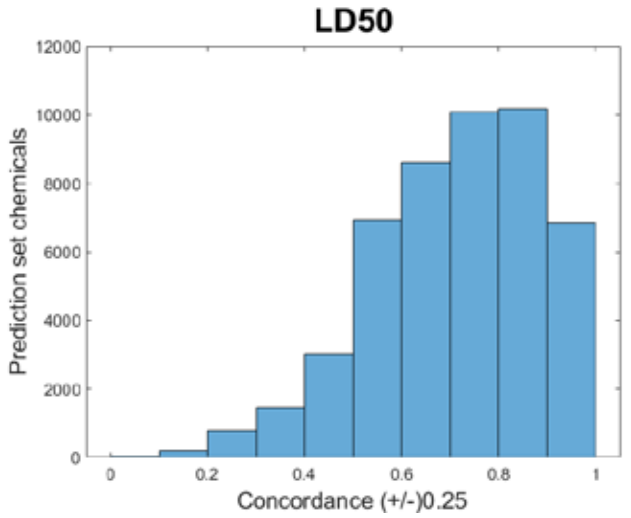
The consensus predictions perform just as well as replicate *in vivo* data do at predicting oral acute toxicity outcome

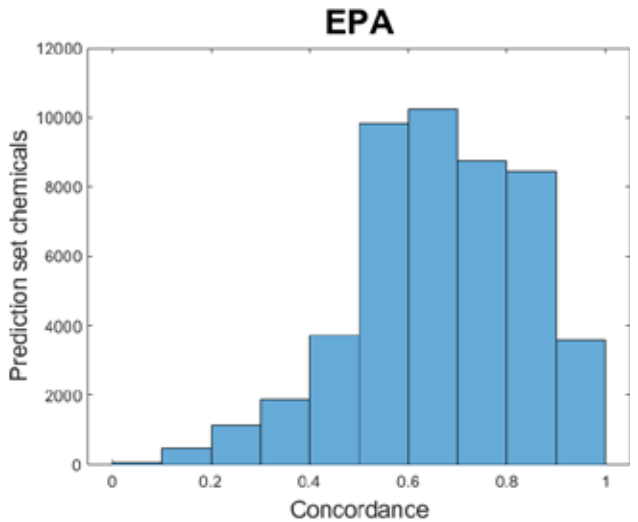
Consensus concordance

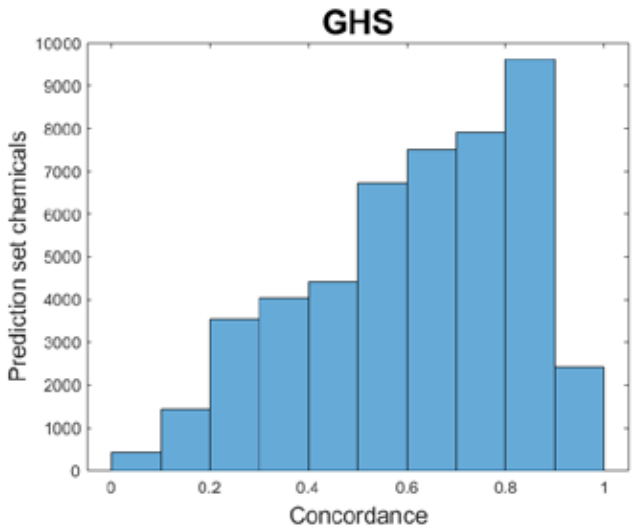
Distributions of the concordance between models











Consensus implementation

Generalized CATMoS models: datasets

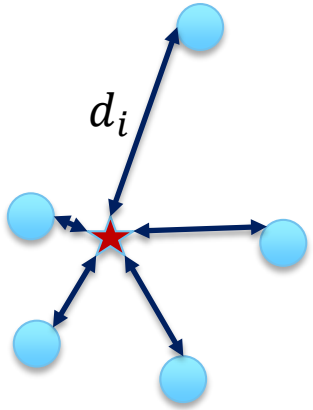
- LD50: 28954
- VT: 23767
- NT: 30971
- EPA: 25487
- GHS: 25720

- High concordance
- Proportional distribution of:
  - LD50 values
  - VT/NT classes
  - EPA/GHS categories

- Split into 75% training and 25% test set
- Calculate PaDEL & CDK2 descriptors
- Dimensionality reduction (missing values & low variance)
- Feature selection (most relevant descriptors for each endpoint)

Consensus implementation

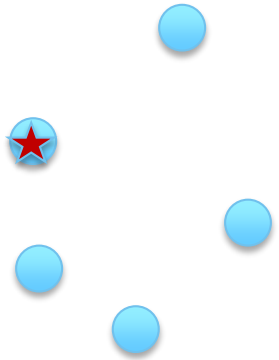
Generalized CATMoS models: new predictions



$d_1 \neq 0$

$w_i = f(d_i)$

$Pred_i = f(w_i, N_i)$



$d_1 = 0$

$Pred_i = N_i$

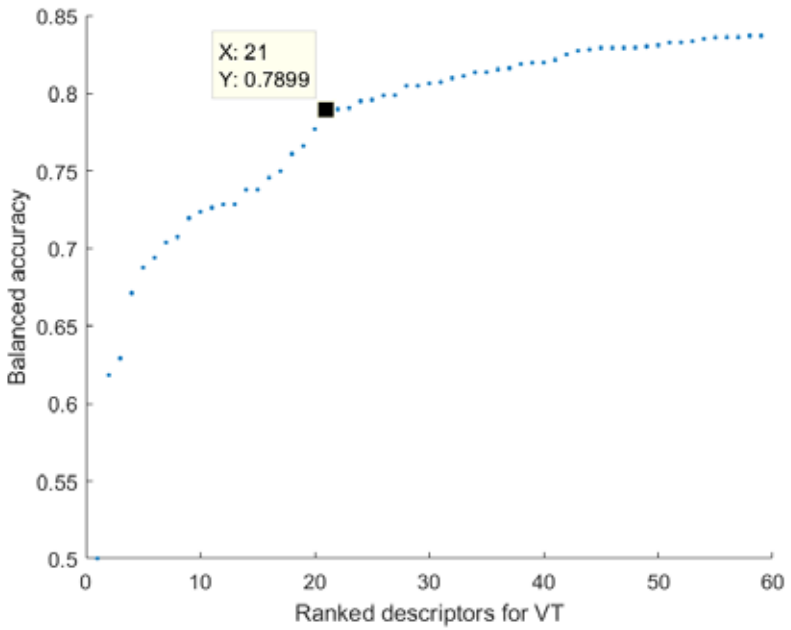
★ New chemical to be predicted ● Nearest neighbors (N_i)

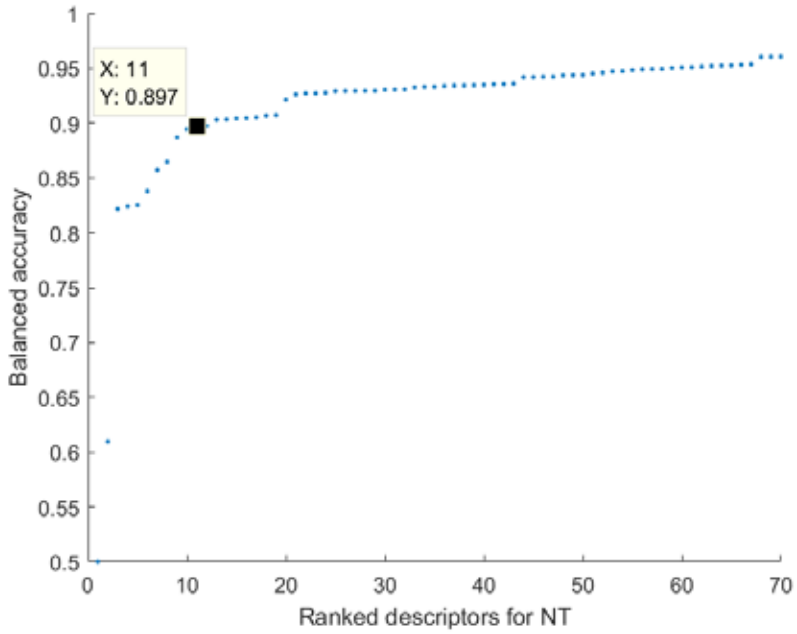
d_i: Euclidean distance based on the selected descriptors for each endpoint

➡ Automated, weighted-endpoint dependent read-across: weighted kNN

Consensus implementation

Generalized CATMoS models: statistics

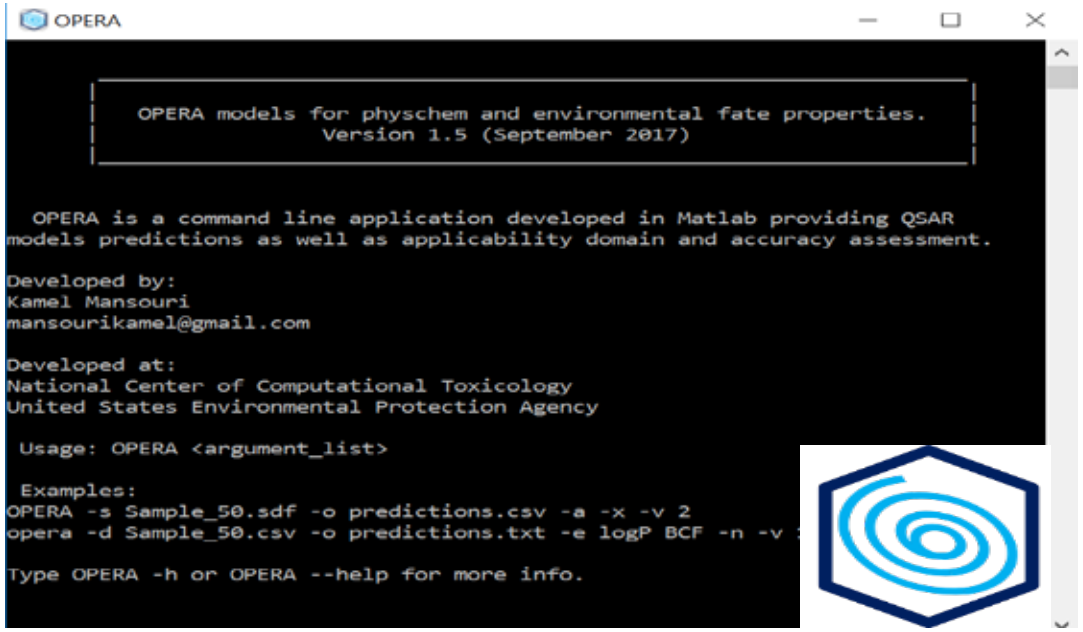




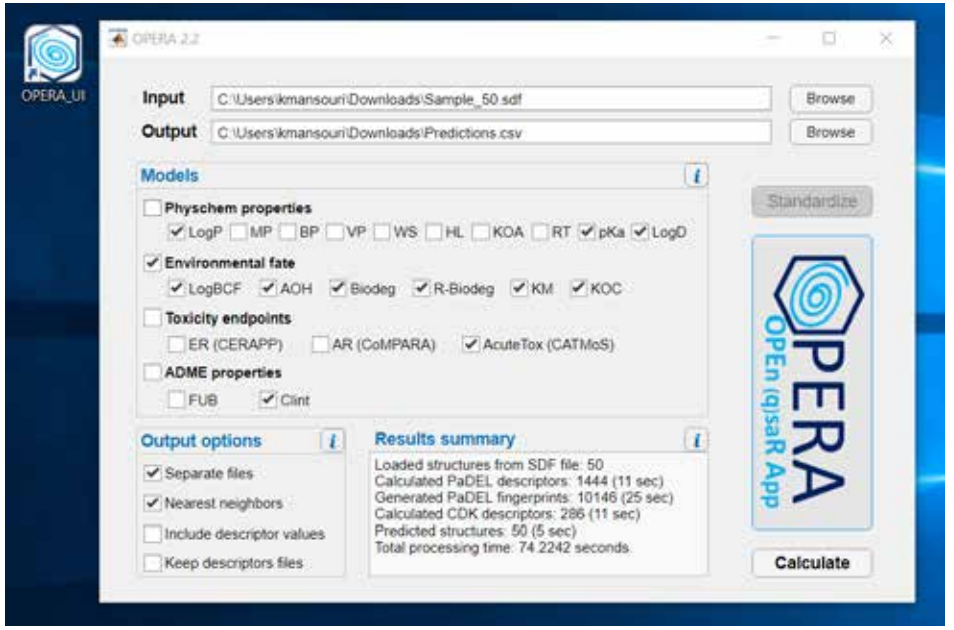
Endpoint	Descriptors	Training (5-f CV)	Test set
VT (BA)	21	0.79	0.77
NT (BA)	11	0.90	0.89
EPA (BA)	15	0.79	0.81
GHS (BA)	15	0.78	0.79
LD50 (Q^2,R^2)	23	0.79	0.81

Access to CATMoS Consensus

OPERA Standalone app



Command line



Graphical user interface

- Free, opensource & open-data
- Single chemical and batch mode
- Multiple platforms (Windows and Linux)
- Embeddable libraries (java, C, C++, Python)

https://github.com/NIEHS/OPERA

Mansouri et al. J Cheminform (2018).
https://doi.org/10.1186/s13321-018-0263-1

OPERA2

OPERA 1.5

Physchem & Environmental fate:

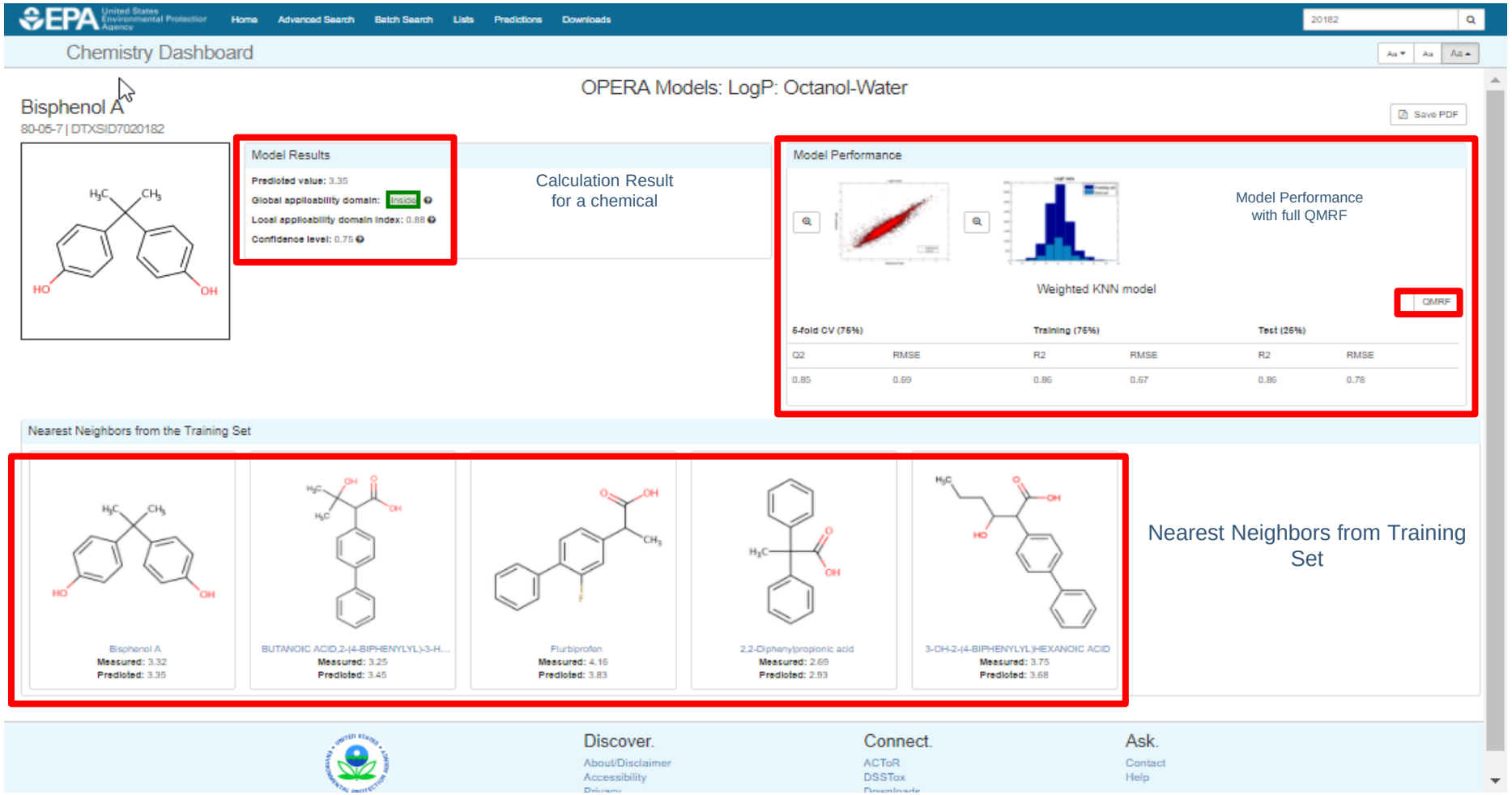
Model	Property
AOH	Atmospheric Hydroxylation Rate
BCF	Bioconcentration Factor
BioHL	Biodegradation Half-life
RB	Ready Biodegradability
BP	Boiling Point
HL	Henry's Law Constant
KM	Fish Biotransformation Half-life
KOA	Octanol/Air Partition Coefficient
LogP	Octanol-water Partition Coefficient
MP	Melting Point
KOC	Soil Adsorption Coefficient
VP	Vapor Pressure
WS	Water solubility
RT	HPLC retention time

New in OPERA2:

- Physchem properties:
  - General structural properties
  - pKa
  - Log D
- Toxicity endpoints
  - ER activity (CERAPP) https://ehp.niehs.nih.gov/15-10267/
  - AR activity (CoMPARA) https://doi.org/10.13140/RG.2.2.19612.80009
  - Acute toxicity (CATMoS) https://doi.org/10.1016/j.comtox.2018.08.002)
- ADME properties
  - Plasma fraction unbound (FuB)
  - Intrinsic clearance (Clint)

OPERA predictions on EPA's CompTox dashboard

https://comptox.epa.gov/dashboard

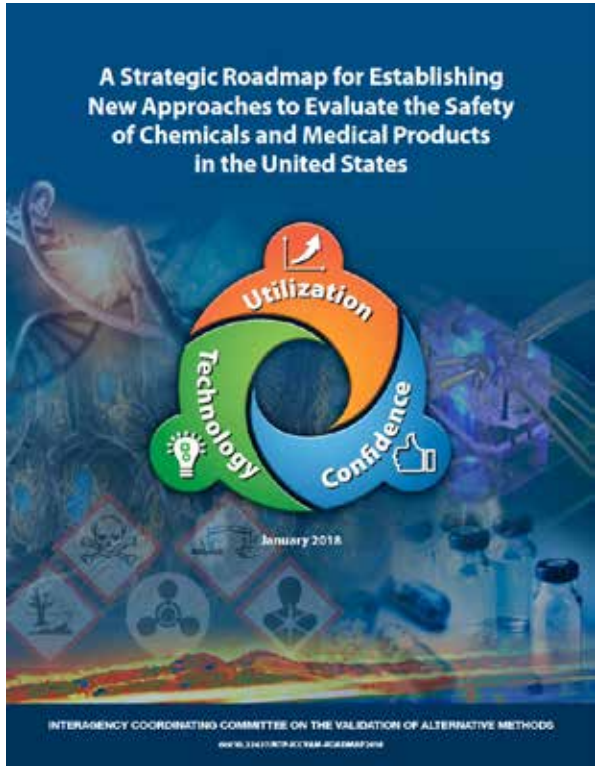


OPERA
OPEn (q)saR App

Mansouri et al. OPERA models (https://doi.org/10.1186/s13321-018-0263-1)
Williams et al. CompTox Chemistry Dashboard (https://doi.org/10.1186/s13321-017-0247-6)

The “3C” Concept at Work!

- Success of the project was due in great part to the use of the 3C concept as well as up-front and continuous engagement of regulators in the process



Communication

Collaboration

Commitment

https://ntp.niehs.nih.gov/go/natl-strategy

Acknowledgements

THANK YOU!

- ICCVAM Acute Toxicity Workgroup

- EPA/NCCT
  - Grace Patlewicz
  - Jeremy Fitzpatrick

- ILS/NICEATM
  - Agnes Karmaus
  - Dave Allen
  - Shannon Bell
  - Patricia Ceger
  - Judy Strickland
  - Amber Daniel

- NTP/NICEATM
  - Nicole Kleinstreuer
  - Warren Casey

Feedback welcome: Kamel Mansouri (kmansouri@ils-inc.com)

Technical support was provided by ILS under NIEHS contract HHSN273201500010C.