

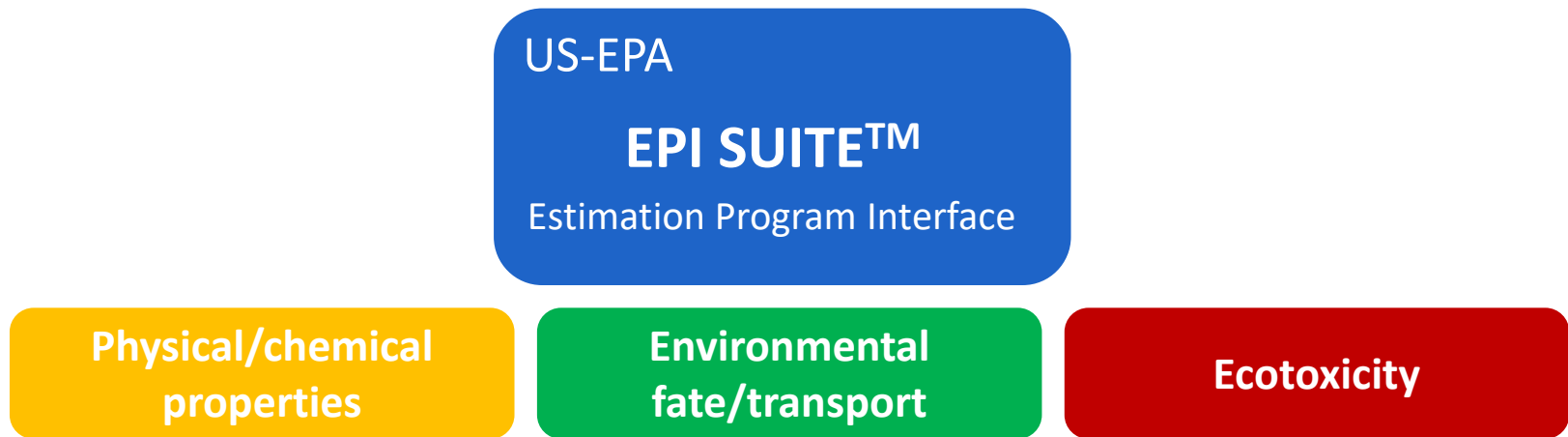
In silico tools for screening of environmental hazard early in the drug development process

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Prof. Colin Janssen and Prof. Jana Asselman

EPI Suite™ – Estimation Program Interface

EPI Suite™ overview



Predictive computerized models

Environmental risk assessment of new chemicals

- Used as a screening level predictive tool
- Screening chemicals for persistence and exposure potential
- Applied for chemicals when the measured values are absent

QSARs available

Physical and chemical properties

KOWWIN™ – logKow

KOCWIN™ – Koc

KOAWIN™ – octanol–air partition coefficient

AOPWIN™ – atmospheric oxidation potential (rate)

HENRYWIN™ – Henry's Law constant

MPBPWIN™ – Melting point, boiling point, and vapor pressure

WSKOWWIN™ – water solubility based on logKow and structural correlation factor

WATERNT™ – water solubility based on fragment approach

Ecotoxicity

ECOSAR: Ecological Structure Activity Relationship

Acute/chronic data for aquatic organisms

US-EPA

EPI SUITE™

Estimation Program Interface

Fate/Transport

AEROWIN™ – atmospheric aerosol formation

BioHCwin – Biodegradation half-life of HC

BCFBAF™ – bioconcentration
HYDROWIN™ – hydrolysis rate

BIOWIN™ – aerobic and anaerobic biodegradability

WVOLWIN™ – rate of volatilization from water

STPWIN™ – removal of a chemical in a sewage treatment plant

LEV3EPI™ – partitioning of chemicals between air, soil, sediment, and water

EPI Suite™ input

The screenshot displays the EPI Suite software interface. At the top, there is a menu bar with options: File, Edit, Functions, Batch Mode, Show Structure, Output, Fugacity, STP, and Help. Below the menu bar is a blue banner with the text "EPI Suite - Welcome Screen".

On the left side, there is a vertical sidebar with a list of program options: AOPWIN, KOWWIN, BIOWIN, MPBPVP, WSKOW, WATERNT, HENRYWIN, KOAWIN, KOCWIN, BCFBAF, HYDROWIN, BioHCwin, DERMWIN, ECOSAR, and EPI Links. Red arrows point from the top of this sidebar towards the main input area.

The main input area contains several fields and buttons:

- Buttons: PhysProp, Previous, Get User, Save User, Search CAS, Calculate (with a calculator icon), Clear Input Fields, Draw.
- Input fields: Input CAS #, Input Smiles, Input Chem Name, Name Lookup.
- Physical and Chemical Properties:
 - Henry LC: atm-m³/mole
 - Water Solubility: mg/L
 - Melting Point: Celsius
 - Vapor Pressure: mm Hg
 - Boiling Point: Celsius
 - Log Kow:
- Environmental Parameters:

	River	Lake	
Water Depth:	<input type="text"/> 1	<input type="text"/> 1	meters
Wind Velocity:	<input type="text"/> 5	<input type="text"/> 0.5	meters/sec
Current Velocity:	<input type="text"/> 1	<input type="text"/> 0.05	meters/sec

On the right side, there is an "Output" section with radio buttons for "Full" and "Summary".

At the bottom of the window, there is a disclaimer text:

The Estimation Programs Interface (EPI) Suite™ was developed by the US Environmental Protection Agency's Office of Pollution Prevention and Toxics and Syracuse Research Corporation (SRC). It is a screening-level tool, intended for use in applications such as to quickly screen chemicals for release potential and "bin" chemicals by priority for future work. Estimated values should not be used when experimental

EPI Suite™ output

The screenshot displays the EPI Suite software interface. At the top, there is a menu bar with options: File, Edit, Functions, Batch Mode, Show Structure, Output, Fugacity, STP, and Help. Below the menu is a blue banner that reads "EPI Suite - Welcome Screen".

On the left side, there is a vertical navigation menu with buttons for various models: AOPWIN, KOWWIN, BIOWIN, MPBPVP, WSKOW, WATERNT, HENRYWIN, KOAWIN, KOCWIN, BCFBAF, HYDROWIN, BioHCwin, DERMWIN, ECOSAR, and EPI Links.

The main area is divided into several sections:

- PhysProp**: A table of physical and chemical properties. The "Molecular Weight" is 270.29 and the "Mol. For." is C15 H14 N2 O3.
- Draw**: A section for drawing the chemical structure, including input fields for CAS #, Smiles, and Chem Name.
- 2D Editor**: A window showing the chemical structure being drawn. The SMILES string is O=C(N)N3c1cccc1C(O)C(O)c2cccc23. The structure is a complex bicyclic molecule with a benzamide group and two hydroxyl groups.

At the bottom of the interface, there is a status bar showing "All Results" and a list of models: KOWWIN, MPBPVP, Water Solubility, ECOSAR, HENRYWIN, KOAWIN, etc. Below this, the CAS Number and SMILES string are displayed.

EPI Suite™ output

Results

All Results | KOWWIN | MPBFVP | Water Solubility | ECOSAR | HENRYWIN | KOAWIN | BIOWIN | BioHCwin | AEROWIN | AOPWIN | KOCWIN | HYDROWIN | BCFBAF | Volatilization | STP

```
CAS Number:  
SMILES : O=C(N)N3c1cccc1C(O)C(O)c2cccc23  
CHEM :  
MOL FOR: C15 H14 N2 O3  
MOL WT : 270.29  
----- EPI SUMMARY (v4.11) -----  
Physical Property Inputs:  
Log Kow (octanol-water): -----  
Boiling Point (deg C) : -----  
Melting Point (deg C) : -----  
Vapor Pressure (mm Hg) : -----  
Water Solubility (mg/L): -----  
Henry LC (atm-m3/mole) : -----  
  
Log Octanol-Water Partition Coef (SRC):  
Log Kow (KOWWIN v1.68 estimate) = -0.21  
  
Boiling Pt, Melting Pt, Vapor Pressure Estimations (MPBFVP v1.43):  
Boiling Pt (deg C): 481.18 (Adapted Stein & Brown method)  
Melting Pt (deg C): 203.81 (Mean or Weighted MP)  
VP (mm Hg, 25 deg C): 3.04E-012 (Modified Grain method)  
VP (Pa, 25 deg C) : 4.05E-010 (Modified Grain method)
```

Create MS Word File | Print Results | Print Results - No Structure | Create a Text file | ISIS Base/Upload TBL File | View Main Screen

Append Data to End of Selected Files

A Note about Creating MS Word files

EPI Suite™ - ECOSAR

Physical and chemical properties

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ECOSAR version 2.0

ECOSAR Application 2.0

ECOSAR Special Cases

Organic Module

Organic

Welcome

ECOSAR Application 2.0

ECOSAR Special Cases

Organic Module

Organic

Organic Module

Chemical Input

Please enter CAS Number or SMILES

Draw

Submit

CAS Number

SMILES

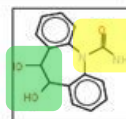
50-00-0, 000050-00-0, ...

O=C

Batch

O=C(N)N3c1cccc1C(O)C(O)c2cccc23

CAS



Log Kow

-0.2068

Water Solubility (mg/L)

103.93

Melting Point (°C)

Chemical Details

SMILES

O=C(N)N3c1cccc1C(O)C(O)c2cccc23

MOL WT

270.29

Log Kow

-0.2068 (estimated)
(measured)

Water Solubility (mg/L)

103.93 (estimated)
(measured)

Organic Module Result

Experimental Data

Physical Properties

K_{ow} Estimate

Report

Substituted Ureas

Organism	Duration	End Point	Concentrati...	Max Log Kow	Flags
Fish	96h	LC50	4.91E+3	5.0	⚠
Daphnid	48h	LC50	4.63E+3	5.0	⚠
Green Algae	96h	EC50	2.57	5.4	
Fish		ChV	87.2	5.0	
Daphnid		ChV	99.8	5.0	
Green Algae		ChV	0.888	5.0	
Fish (SW)	96h	LC50	263	5.0	⚠
Mysid (SW)	96h	LC50	867	5.0	⚠
Fish (SW)		ChV	7.96	5.0	
Mysid (SW)		ChV	682	5.0	⚠

Benzyl Alcohols

Organism	Duration	End Point	Concentrati...	Max Log Kow	Flags
Fish	96h	LC50	7.21E+3	5.0	⚠
Daphnid	48h	LC50	4.81E+3	5.0	⚠
Green Algae	96h	EC50	899	5.4	⚠
Fish		ChV	398	5.0	⚠
Daphnid		ChV	458	5.0	⚠
Green Algae		ChV	252	5.0	⚠



ECOSAR Version 2.0



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Disclaimer: Experimental data sources and values estimated by EPI are not endorsed by the EPA; nor do they represent the EPA's position. Furthermore, professional judgement is needed to determine the applicability and accuracy of Physical Properties.

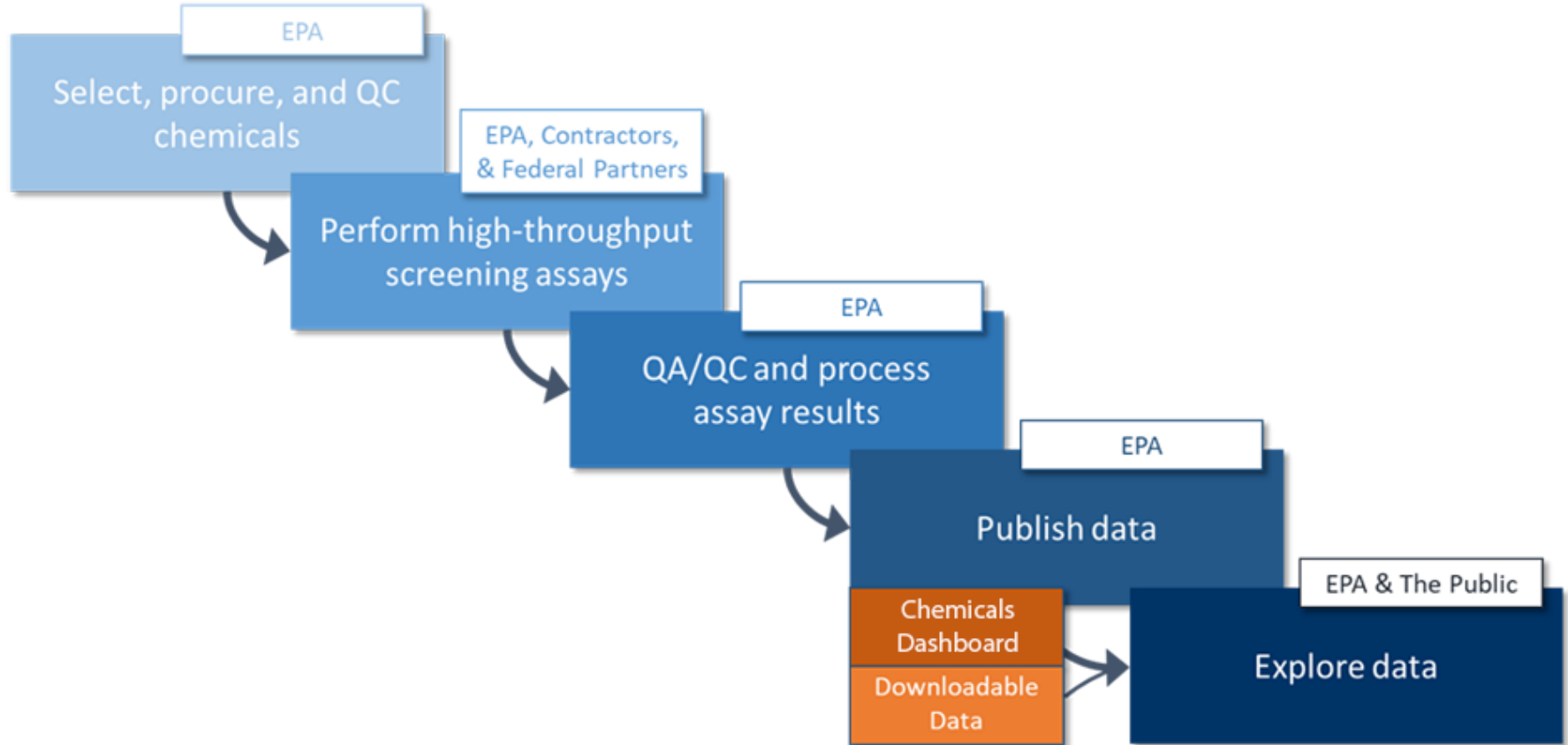
Accept

Decline

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ToxCast database

ToxCast high-throughput database



Potential applications include

- Indication of toxicity range - using similar chemicals
- Identification of most suitable assays
- Prioritization of chemicals for further testing

ToxCast chemical selection criteria



Relevance

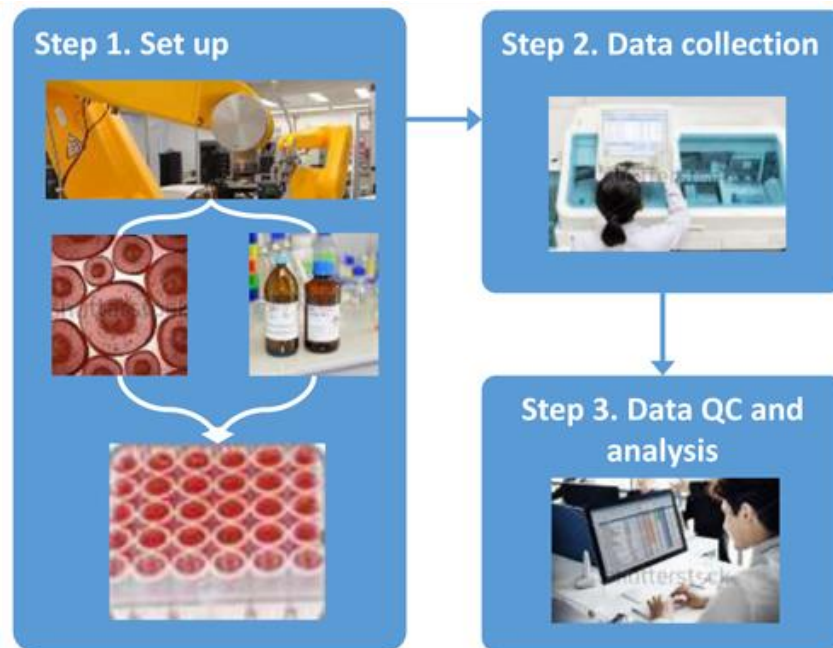
- Exposure potential
- Regulatory interest



Practical constrains

- Commercial availability
- DMSO solubility
- Volatility
- Chemical stability

Generating ToxCast data



Step 1.

High-throughput technology set-up assays into plates with numerous wells (e.g. 384 well-plate)

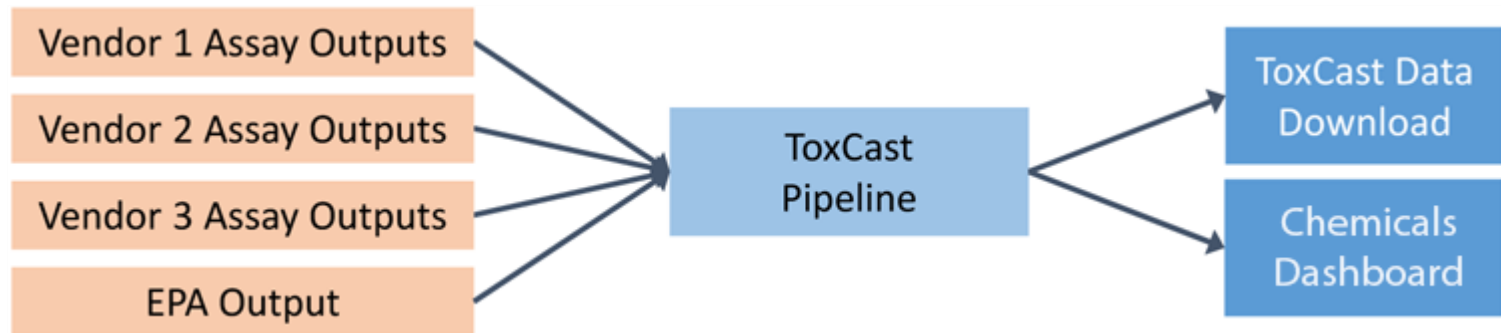
Step 2.

Incubation, assay performance and data compilation

Step 3.

Quality control, normalization, analysis and formatting

Data processing and analysis



Data compilation from different sources in unique formats

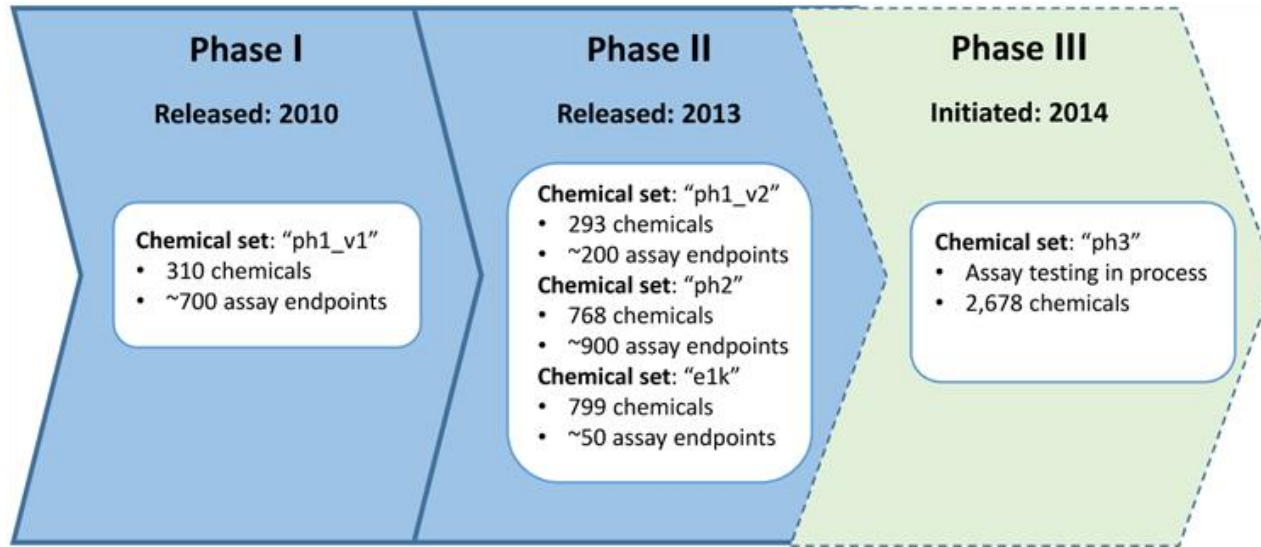


Data upload into ToxCast Pipeline for processing, modeling, inspection and visualization



Final data made available for download and in the Chemicals Dashboard

Publicly available data

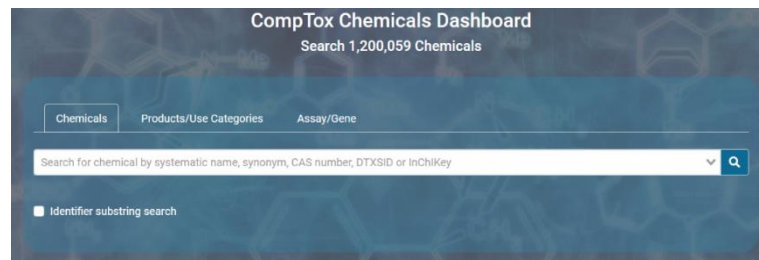


➔ ToxCast has been developed in three distinct phases

➔ Currently data available for ~ 1800 chemicals and 700 assay endpoints

➔ Within Phase III 2678 more chemicals will be tested

Accessing ToxCast data



<https://comptox.epa.gov/dashboard/>



[https://epa.figshare.com/articles/dataset/
ToxCast_Database_invitroDB_/6062623](https://epa.figshare.com/articles/dataset/ToxCast_Database_invitroDB_/6062623)

Accessing ToxCast data

Accession via

Chemical information

- systematic name
- synonym
- CASRN
- DTXSID – DSSTox substance identifier
- InChIKey – International Chemical Identifier

Assay/Gene information

- endpoint name
- target gene name or symbol

Data available on

tested assays – including assay details

target protein ID – connection to SeqAPASS

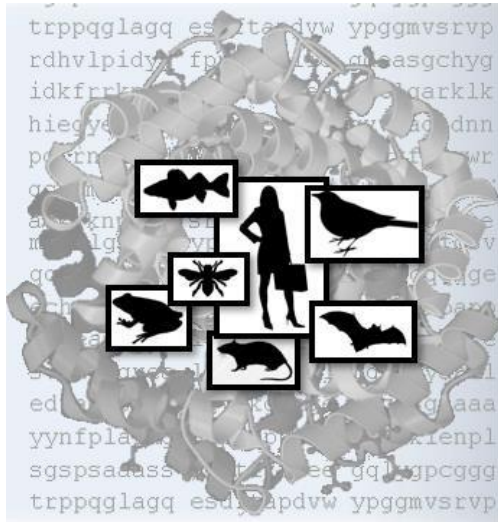
target gene ID

connection to AOP-Wiki

toxicity data expressed as AC50 (μM) - 50% of maximum activity

SeqAPASS – Sequence Alignment to Predict Across Species Susceptibility

What is SeqAPASS?



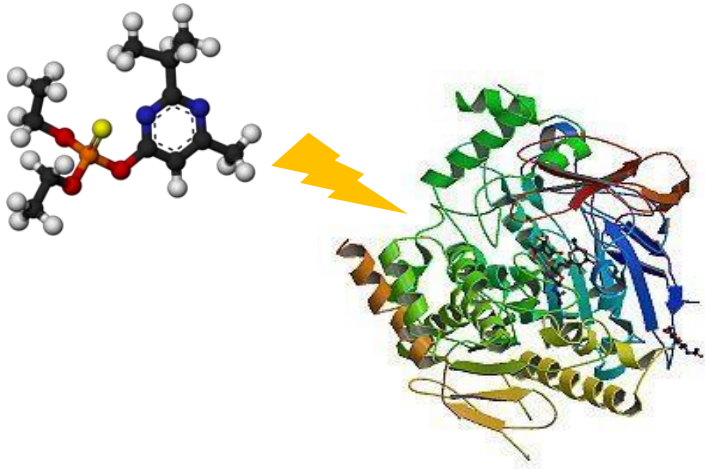
Sequence Alignment to Predict Across Species Susceptibility - SeqAPASS

Uses protein sequence alignment information

Evaluates sequence similarity across species

Predicts species susceptibility

Required information



➔ Knowledge of sensitive or targeted species
– targeted protein

➔ Protein sequence – e.g. NCBI

Sequence of chemical molecular target in target species



Compared to **millions of proteins**
from **thousands of species**

**Greater similarity = greater likelihood of
chemical interaction**

Multi-level analysis

Level 1


Primary Amino Acid Sequence Alignments

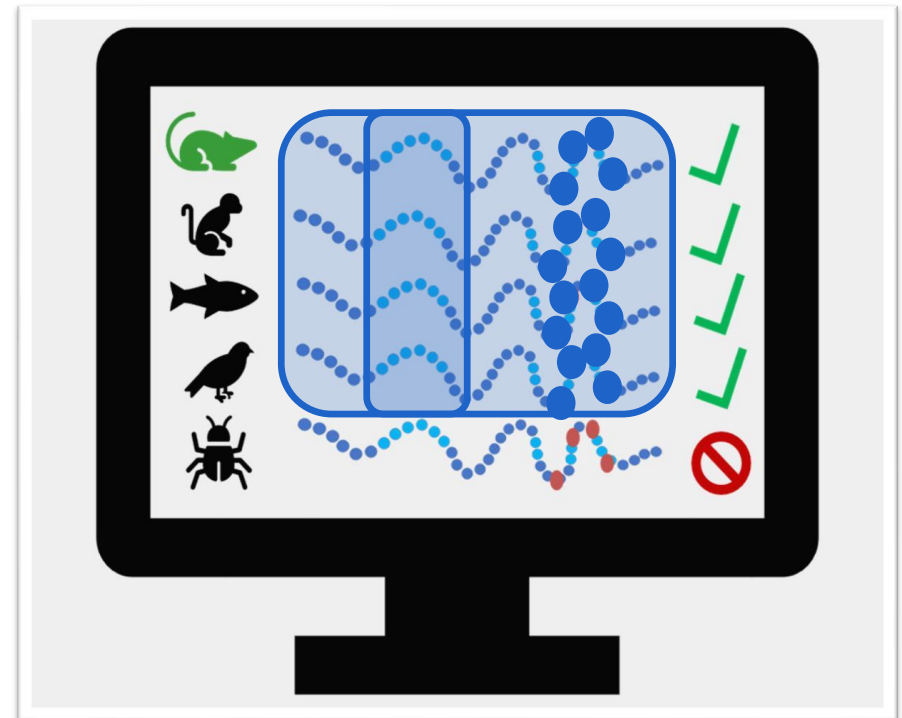
Level 2

Conserved Functional Domain Alignments – e.g. DNA binding domain

Level 3

Critical Amino Acid Conservation

 **Gathers lines of evidence of protein conservation across species**



SeqAPASS output

- ➔ **Percent similarity** is quantified
- ➔ **Susceptibility cut-off** is estimated
- ➔ **Qualitative output (YES/NO)** based on protein conservation across different levels for various species

Common Name	Ortholog Candidate	Cut-off	Percent Similarity
Human	Y	33.15	100
Florida manatee	Y	33.15	98.8
Mallard	Y	33.15	82.29
Rock pigeon	Y	33.15	80.93
Green anole	Y	33.15	80.65
Pacific transparent sea squirt	Y	33.15	33.15
Yesso scallop	N	33.15	32.87
Purple sea urchin	N	33.15	26.05
Human whipworm	N	33.15	23.53
Bed bug	N	33.15	21.62

A blue arrow points down from the 'Percent Similarity' column header to the value 33.15 for the Pacific transparent sea squirt. A red dashed horizontal line is drawn across the table at the 33.15 similarity level. A green box highlights the row for Pacific transparent sea squirt, and a text label 'Lowest % Similarity that is still an ortholog' is positioned to the right of the table.

Other interesting tools

- OECD QSAR Toolbox - <https://www.oecd.org/chemicalsafety/risk-assessment/oecd-qsar-toolbox.htm>
- VEGA QSAR - <https://www.vegahub.eu/portfolio-item/vega-qsar/>

Both include human health hazard assessment

- ECOdrug - <https://ecodrug.org/>

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