EPA's DSSTox Chemical Database: *A Resource for the Non-Targeted Testing Community*

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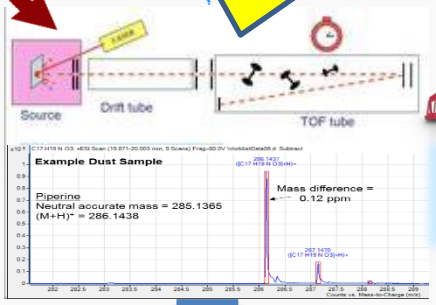
Outline

- Cheminformatics view of problem
- DSSTox chemical database
- ToxCast chemical library
- Tox21 analytical QC
- Challenges

Cheminformatics view of non-targeted testing problem



Thousands of mass ion peaks & abundances

What's in the sample?
&
Should I be worried?

{mono-isotopic mass}

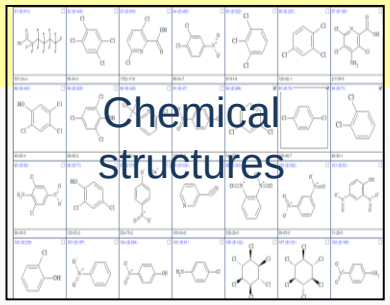
MI mass

1:many

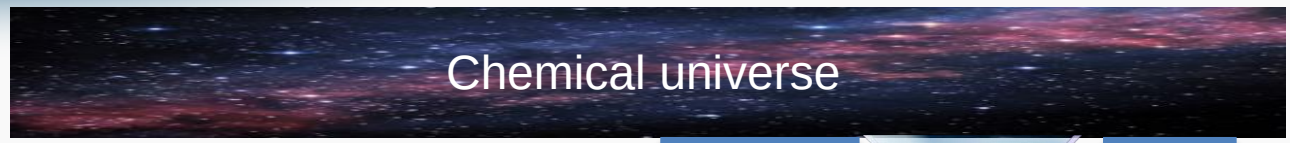
{formulas}

1:many

{parent structures}



structure



Exposure?

Toxic?

Suspect testing, physical standards

Chemical substance database

Inventory/Data

CURATION

{CAS-name}

CURATION

1:1

CURATION

{structure}

How big is the problem?

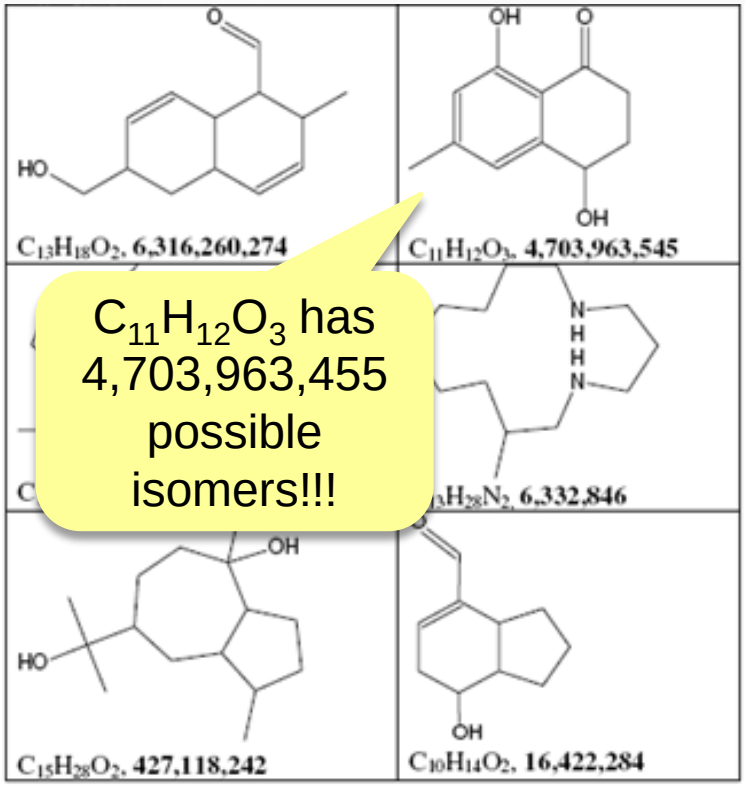
http://www.chemspider.com/blog/

03 02 2008

How Many Structures Can You Generate From A Molecular Formula?

Posted by: Antony Williams in ChemSpider Chemistry *(22 million ChemSpider IDs in 2008)*

Theoretically…



$C_{11}H_{12}O_3$ has 4,703,963,455 possible isomers!!!

Highest frequency formula ChemSpider?

ID	Structure	Empirical Formula	Molecular Weight	Monoisotopic Mass, Da
33808	load save zoom jmol	$C_{18}H_{20}N_2O_3$	312.363	312.147393
81525	load save zoom jmol	$C_{18}H_{20}N_2O_3$		
103086	load save zoom jmol	$C_{18}H_{20}N_2O_3$	312.363	312.147393

$C_{18}H_{20}N_2O_3$ occurs 5110 times

How big is the problem?

http://www.chemspider.com/blog/

How Many Structures Can You Generate From A Molecular Formula?

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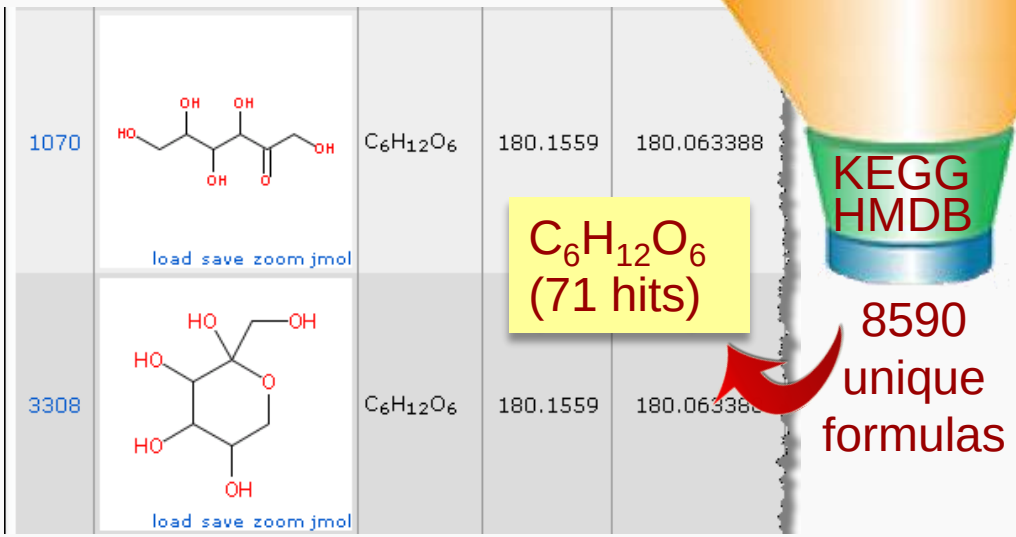
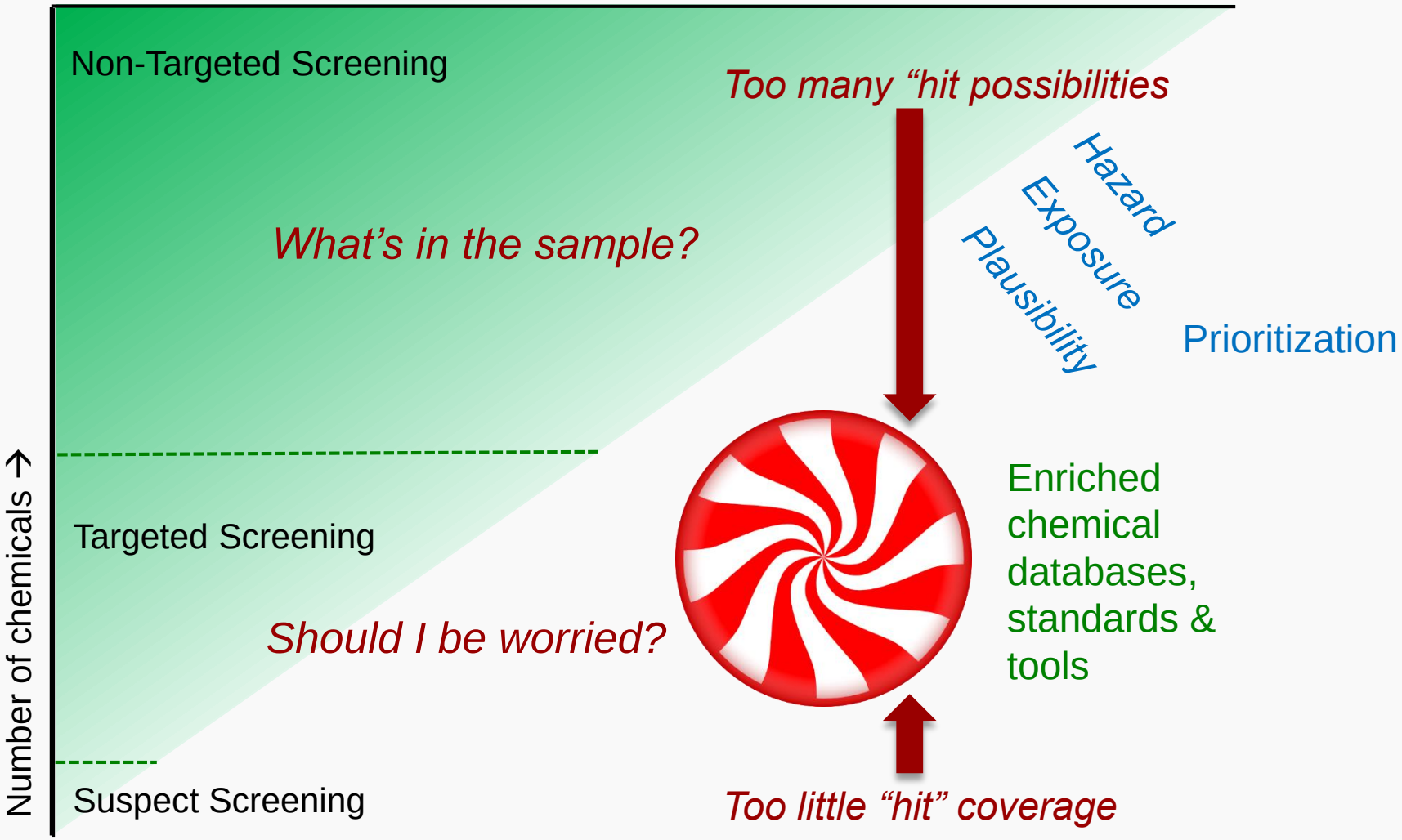


Problem too big to solve with formula matching & structure enumeration

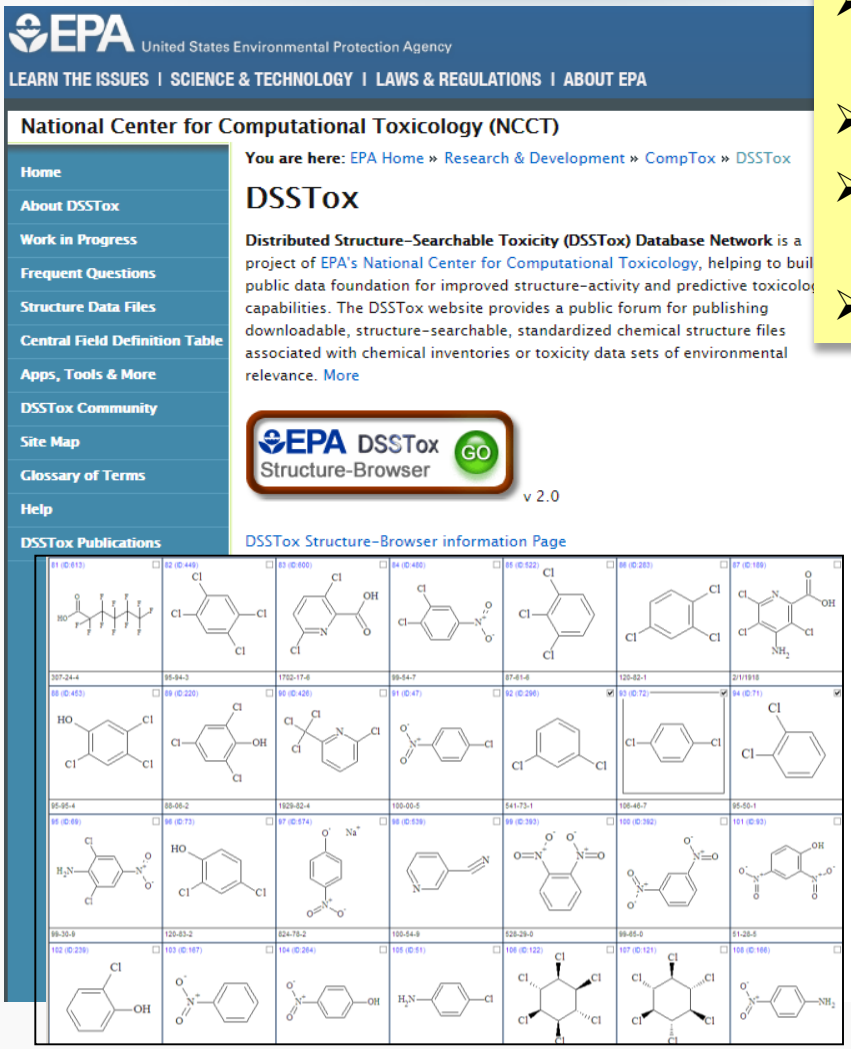
Cheminformatics view of non-targeted testing problem



DSSTox_v1 (thru 3/2014)

http://www.epa.gov/ncct/dsstox/

- Original target audience: Structure-Activity Relationship (SAR) toxicity modeling community
- Focus on EPA, HPV, environmental toxicity datasets
- Emphasis on accurate CAS-name-structure annotations at substance level
- Public resource for high-quality structure-data files



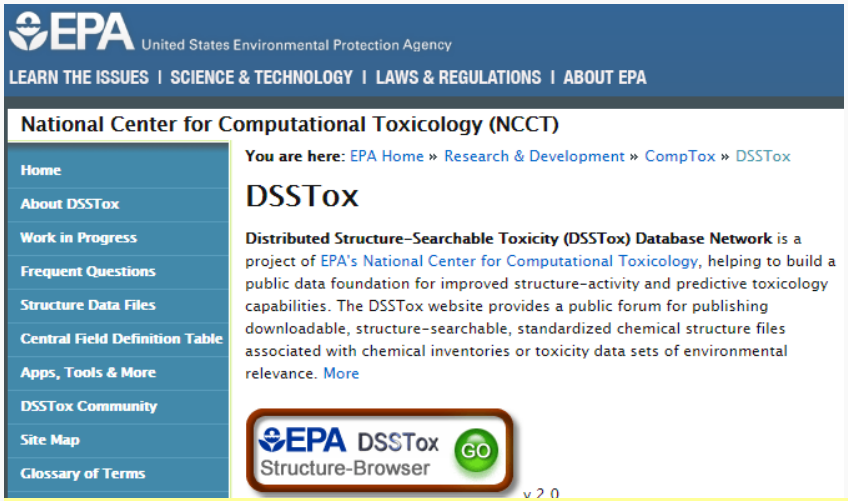
- ARYEXP_v2a_958_06Mar2009
- CPDBAS_v5d_1547_20Nov2008
- DBPCAN_v4b_209_15Feb2008
- EPAFHM_v4b_617_15Feb2008
- FDAMDD_v3b_1216_15Feb2008
- GEOGSE_v2a_1179_09Mar2009
- HPVCSI_v2c_3548_15Feb2008
- HPVISD_v1b_1006_15Feb2008
- IRISTR_v1b_544_15Feb2008
- KIERBL_v1a_278_17Feb2009
- NCTRER_v4b_232_15Feb2008
- NTPBSI_v4c_2330_04Aug2009
- NTPHTS_v2c_1408_11Mar2009
- TOX21S_v2a_8193_22Mar2012*
- TOXCST_v4a_1892_20Mar2012*
- External Data Files (ISSCAN)

DSSTox_RID
DSSTox_GSID
DSSTox_CID
DSSTox_FileID
TestSubstance_ChemicalName
TestSubstance_CASRN
TestSubstance_Description
ChemicalNote
STRUCTURE_Shown
STRUCTURE_Formula
STRUCTURE_MolecularWeight
STRUCTURE_ChemicalType
STRUCTURE_TestedForm_DefinedOrganic
STRUCTURE_ChemicalName_IUPAC
STRUCTURE_SMILES
STRUCTURE_Parent_SMILES
STRUCTURE_InChIS
STRUCTURE_InChIKey
Substance_modify_yyyymmdd

Approx. 25K CAS-substances, 16K structures

DSSTox Update

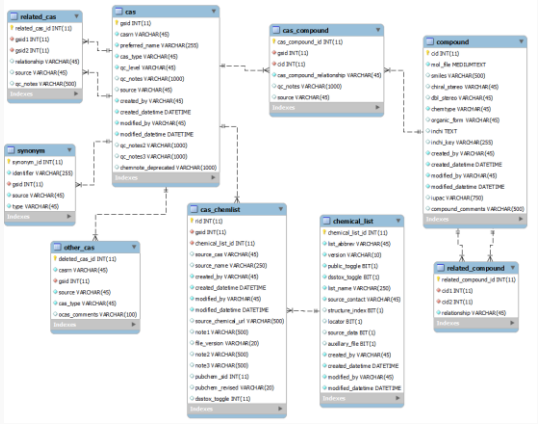
DSSTox_v1



- Website to be retired 9/30/2015
- DSSTox_v1 files & documentation will remain available on EPA ftp site
- Original 25K substance records stored in MS ACCESS and Excel tables serve as the initial Level 1,2 input to DSSTox_v2

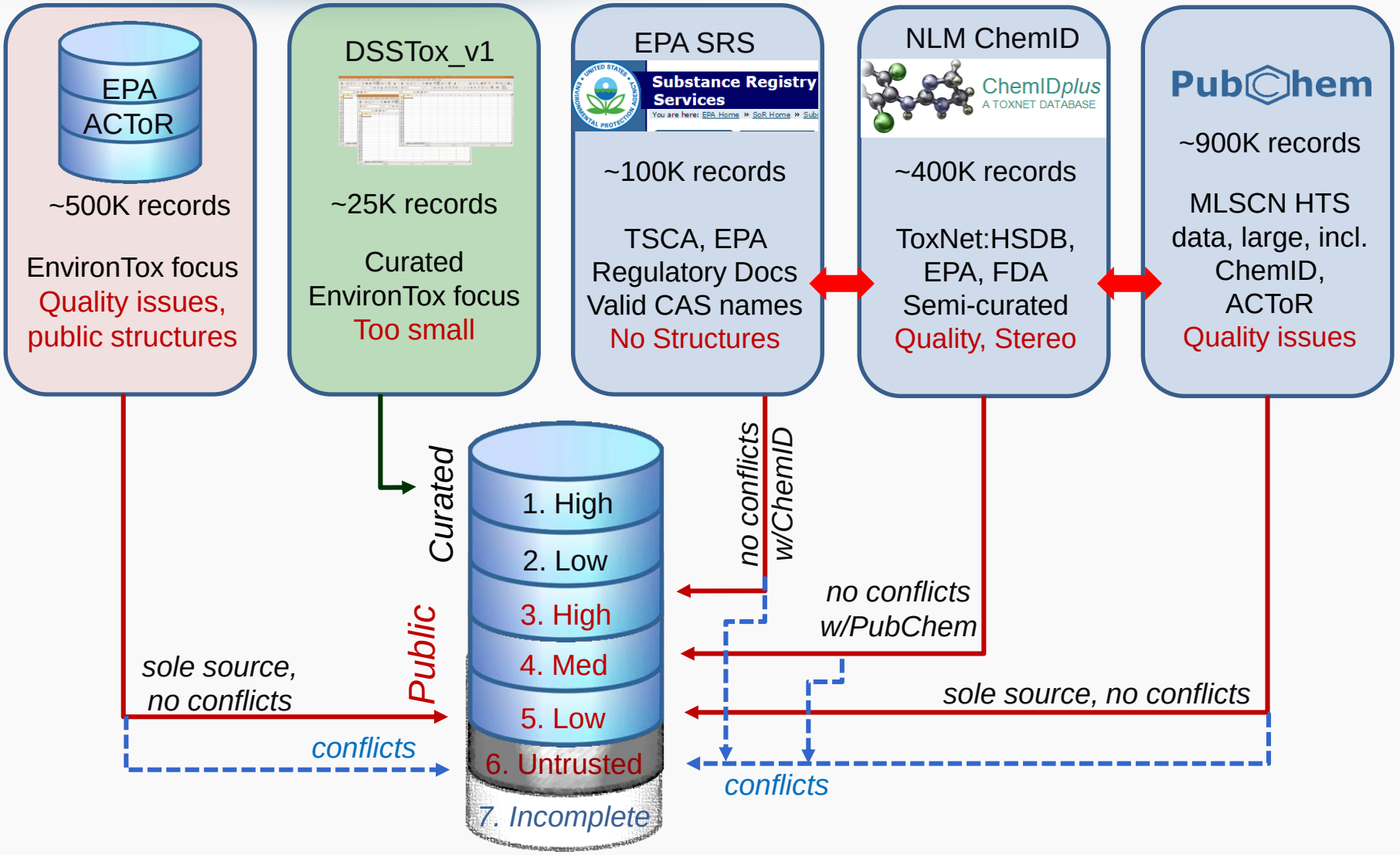
DSSTox_v2

- Convert DSSTox tables to MySQL
- Develop curation interface
- Implement cheminformatics workflow
- Expand chemical content
- Register ACToR data inventories
- Web-services & Dashboard access

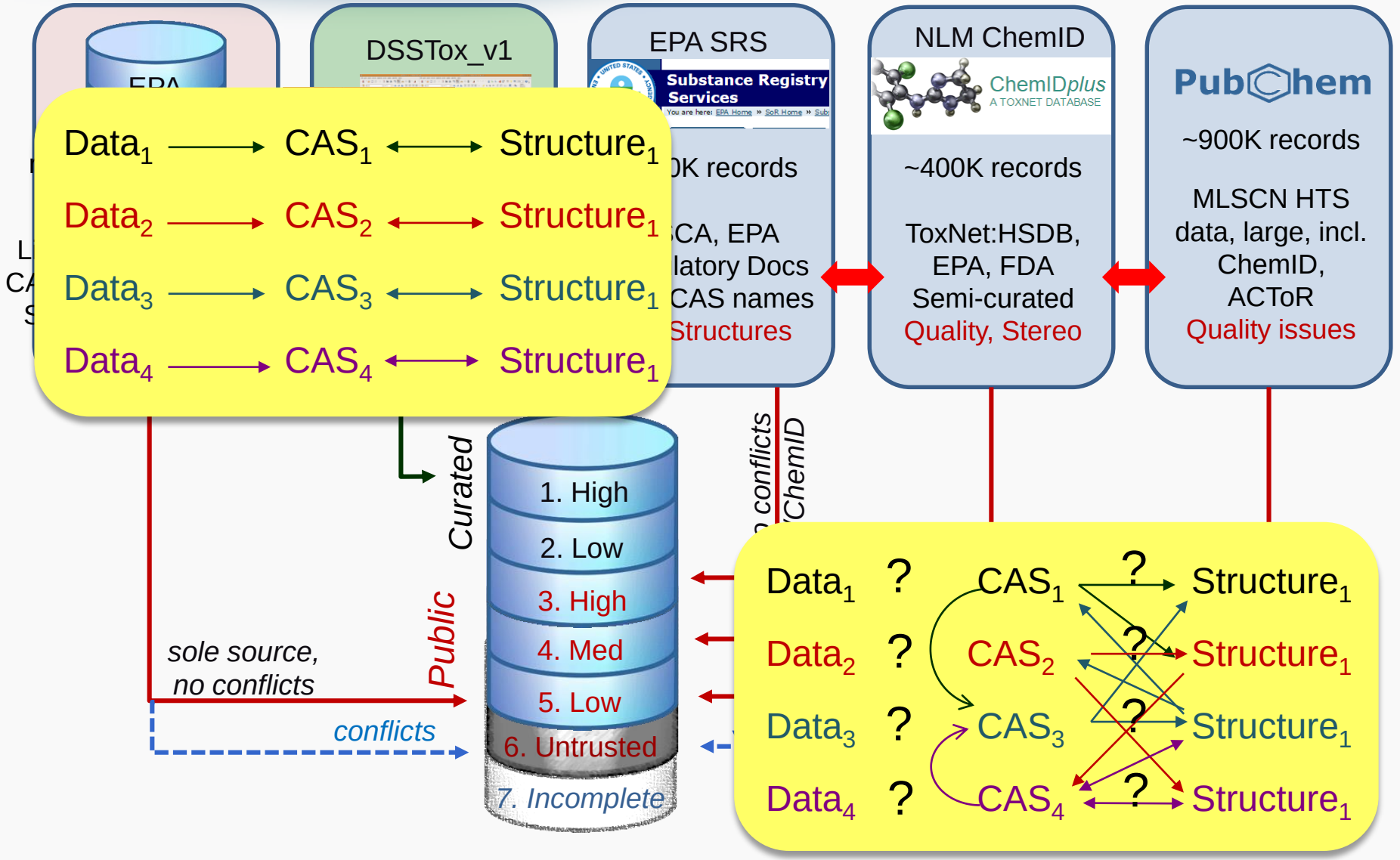


DSSTox_v2 CAS-Structure Sources: QC levels

EPA United States Environmental Protection Agency



DSSTox_v2 CAS-Structure Sources: QC levels



DSSTox_v2 Construction

Data source load order:

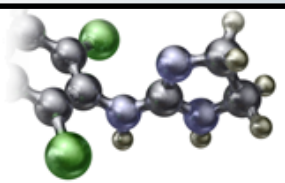
1) DSSTox_v1 (~22K)
   - ✓ 1:1 CAS-structure mappings
   - ✓ Assign NOCAS_GSID
   - ✓ Related CAS & structure mappings (e.g., NOCAS, mixtures)

2) EPA SRS (~77K)
   - ✓ *systematic name → structure conversion*
   - ✓ *internal CAS-structure conflicts (12.5%)*
   - ✓ *ChemID conflicts (24% of 30K overlaps)*
   - ✓ *DSSTox conflicts (8% of 6200 overlaps) → queue for curation*

3) ChemID (~77K)
   - ✓ *internal CAS-structure conflicts (4.5%)*
   - ✓ *PubChem conflicts (45% of 225K overlaps) … OUCH!!*
   - ✓ *DSSTox conflicts (11% of 2300 overlaps) → queue for curation*

4) And so on …

Example problem



ChemIDplus
A TOXNET DATABASE

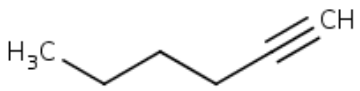
Start New Query | Modify Query | Search History | Show Query

Switch to Summary View

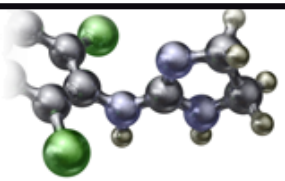
Substance Name: Hexyne
RN: 26856-30-4
InChIKey: CGHIBGNXEGJPQZ-UHFFFAOYSA-N

Molecular Formula
C6-H10

Molecular Weight
82.145



Na +



ChemIDplus
A TOXNET DATABASE

Start New Query | Modify Query | Search History | Show Query

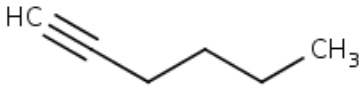
Switch to Summary View

Go to summary view

Substance Name: 1-Hexyne
RN: 693-02-7
InChIKey: CGHIBGNXEGJPQZ-UHFFFAOYSA-N

Molecular Formula
C6-H10

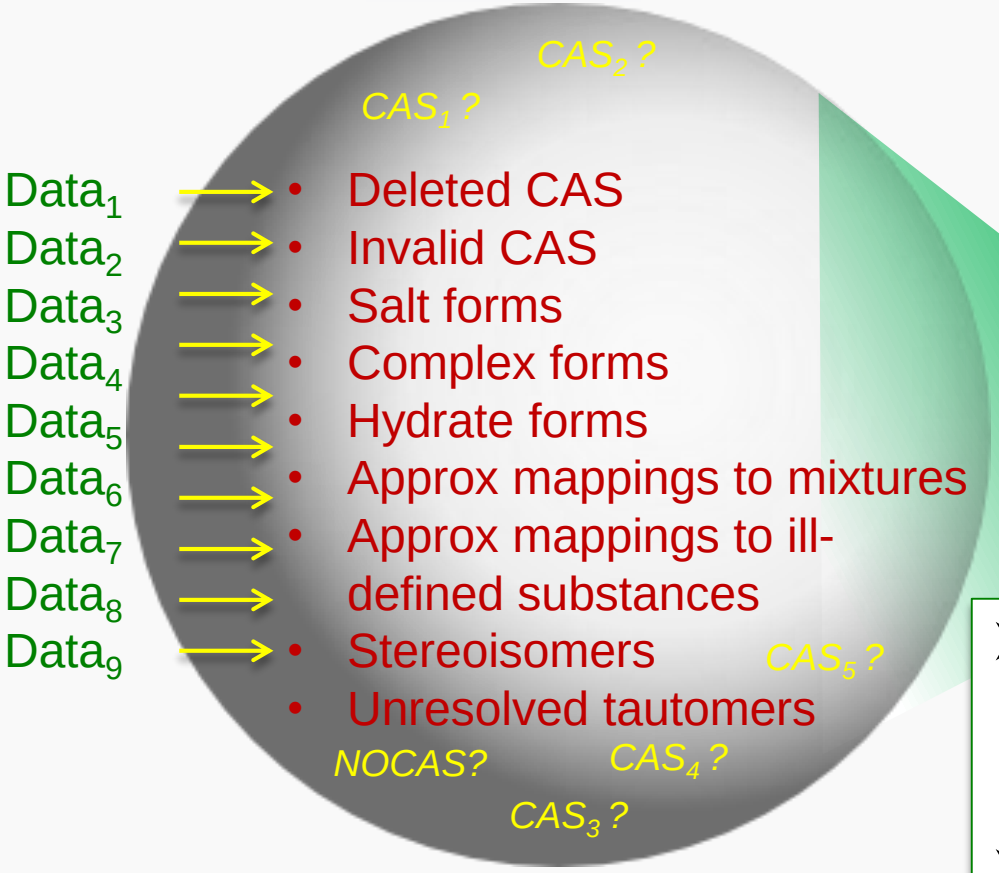
Molecular Weight
82.145



Na +

3D

CAS-Structure "Sphere of Confusion"



many:1

Monoisotopic Mass

↓

Formula

↓

Parent structure
(no stereo, desalted)

↓

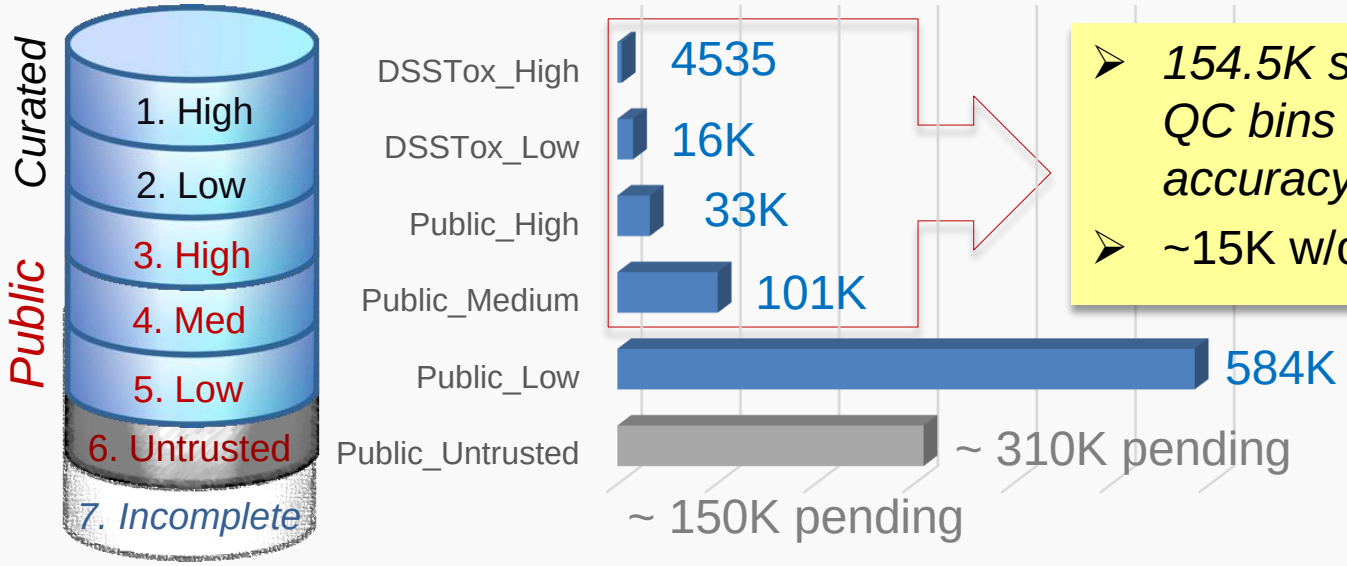
Valid CAS-substance?

- Resolve CAS-structure mappings for accurate chemical-data mapping, i.e. what was tested?
- Collapse sphere to bring all related data to NTS parent structure-formula level

DSSTox_v2 Database & Cheminformatics Layer

DSSTox_v2 Totals

QC Level Totals (12Jun2015)



- *154.5K substances in top 4 QC bins exceed public accuracy standards*
- ~15K w/o structures

QC Levels

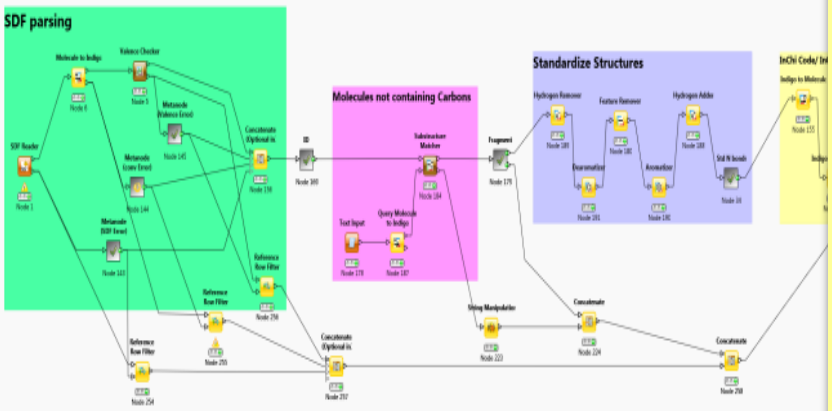
DSSTox_High:	Hand curated and validated
DSSTox_Low:	Hand curated and confirmed using multiple public sources
Public_High:	Extracted from EPA SRS and confirmed to have no conflicts in ChemID and PubChem
Public_Medium:	Extracted from ChemID and confirmed to have no conflicts in PubChem
Public_Low:	Extracted from ACToR or PubChem
Public_Untrusted:	Postulated, but found to have conflicts in public sources

KNIME structure-“cleaning” workflow

https://www.knime.org/knime

Objectives:

- Combine community approaches to structure processing
- Develop a flexible workflow to be used by EPA and shared publicly
- Process DSSTox files to create “QSAR-ready” structures



- ✓ Parse SDF, remove fragments
- ✓ Explicit hydrogen removed
- ✓ Dearomatization
- ✓ Removal of chirality info, isotopes and pseudo-atoms
- ✓ Aromatization + add explicit hydrogen atoms
- ✓ Standardize Nitro groups
- ✓ Other tautomerize/mesomerization
- ✓ Neutralize (when possible)

Publicly available cheminformatics toolkits in KNIME:

Indigo

RDKit
Open-Source Cheminformatics and Machine Learning

Pharmaceutical Data Exploration Laboratory

Slide courtesy of K. Mansouri

DSSTox Viewer (EPA Intranet)

EPA United States Environmental Protection Agency

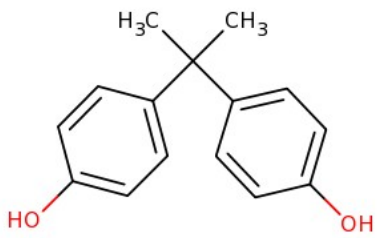
http://rtpavaki1.epa.gov:8080/DSSToxViewer/

DSSTox Viewer

Search a Single Record

Welcome
Matched Name.
You are viewing the record associated with
GSID: 20182
CASRN: 80-05-7

GSID
or
CAS
or
Name bisphenol A
search



GSID: 20182
CAS: 80-05-7
Name: Bisphenol A
Substance Type: Single Compound
QC Level: DSSTox_High

QC Notes:

CID:
Chemica
Shown:
CAS-Co
Relation
Notes:
Source
Compou
Double Stereo:
Chiral Stereo:
Chemical Form: Organic

Synonyms (1)

Synonym	Source
Bisphenol A (BPA) (4,4′-Propane-2,2-diyldiphenol) (Phenol, 4,4'-(1-methylethylidene)bis-)	DSSTox

Other Cas (5)

CAS-RN	Relationship	
137885-53-1	Deleted	SRS
146479-75-6	Deleted	SRS
27360-89-0	Deleted	SRS
28106-82-3	Deleted	SRS
37808-08-5		

- *35% DSSTox_High have deleted CAS*
- *>20K total deleted CAS*

QC Levels

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DSSTox_Low:	Hand curated and confirmed using multiple public sources
Public_High:	Extracted from EPA SRS and confirmed to have no conflicts in ChemID and PubChem
Public_Medium:	Extracted from ChemID and confirmed to have no conflicts in PubChem
Public_Low:	Extracted from ACToR or PubChem
Public_Untrusted:	Postulated, but found to have conflicts in public sources

DSSTox Batch Tool (EPA Intranet)

http://rtpavaki1.epa.gov:8080/DSSToxViewer/

DSSTox Viewer

Search a Single Record | Export DSSTox

Choose your identifier type

Name

Submit a list of Identifiers

```
Ecolyst
Valifenalate
Empenthrin [(EZ)- (1R)-
isomers]
Pyrafluprole
Fluoxastrobin
Pyriprole
Benthiavalicarb
Pyribencarb
Triclopyricarb
Meperfluthrin
Pyrametostrobin
Heptafluthrin
Zeta-Cypermethrin
```

Download DSSTox Gsids

Download DSSTox Structures

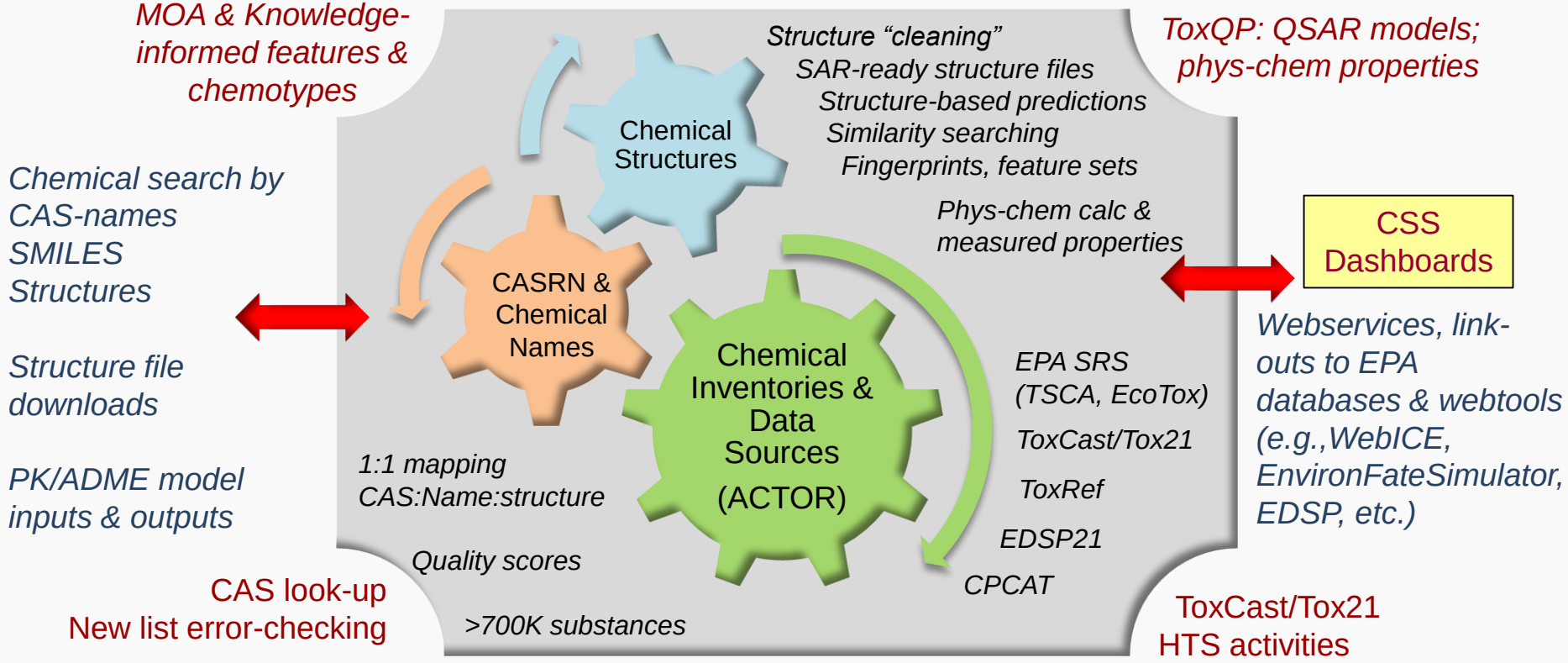
Paste | Calibri | 11 | Wrap Text | General | Conditional Formatting | Clipboard | Font | Alignment | Number | Style

B2 | Matched Name.

	A	B	C	D	E	F	G	H	I	J
1	Query	Found_By	DSSTox_G	DSSTox_C	ChemNan	CASRN	QC_Level	Descriptic	ChemNot	STRUCTU
2	Methoxyfenozide	Matched Name.	32628	12628	Methoxyf	161050-58	DSSTox_L	Single Compound		C22H28N
3	Fenamidone	Matched Name.	34590	14590	Fenamido	161326-34	DSSTox_L	Single Compound		C17H17N
4	(S)-Dimethenamid	Not Found								
5	Selamectin	Matched Name.	45903	25903	Selamecti	165108-07	DSSTox_L	Single Cor	tautomers	C43H63N
6	Dinotefuran	Matched Name.	34549	14549	Dinotefur	165252-70	DSSTox_L	Single Compound		C7H14N4
7	Lepimectin	Matched Name.	58236	32043	Lepimecti	171249-05	DSSTox_H	Single Cor	Absolute	C41H53N
8	Indoxacarb	Matched Name.	32690	12690	Indoxacar	173584-44	DSSTox_H	Single Compound		C22H17C
9	Pyraclostrobin	Matched Name.	32638	12638	Pyraclostr	175013-18	DSSTox_L	Single Compound		C19H18C
10	Ethiprole	Matched Name.	58003	31771	Ethiprole	181587-01	DSSTox_H	Single Compound		C13H9Cl
11	Acetoprole	Matched Name.	58017	31785	Acetoprol	209861-58	DSSTox_H	Single Compound		C13H10C
12	Clothianidin	Matched Name.	34465	14465	Clothianic	210880-92	DSSTox_L	Single Cor	tautomers	C6H8ClN
13	Profluthrin	Matched Name.	58023	31791	Profluthri	223419-20	DSSTox_H	Single Compound		C17H18F
14	Cloflubicyne	Matched Name.	274166	196562	Cloflubicy	224790-70	DSSTox_L	Single Cor		C11H6Cl
15	Metofluthrin	Matched Name.	34738	14738	Metofluth	240494-70	DSSTox_L	Single Compound		C18H20F
16	Pinoxaden	Matched Name.	34823	14823	Pinoxader	243973-20	DSSTox_L	Single Compound		C23H32N
17	Dimefluthrin	Matched Name.	58043	31811	Dimefluth	271241-14	DSSTox_H	Single Compound		C19H22F
18	Ecolyst	Not Found								
19	Valifenalate	Matched Name.	58051	31819	Valifenala	283159-90	DSSTox_H	Single Cor	Absolute	C19H27C
20	Empenthrin [(EZ)- (1R	Matched Name.	58243		Empenthr	303021-82	DSSTox_H	Mixture/F	Absolute	stereoche
21	Pyrafluprole	Not Found								

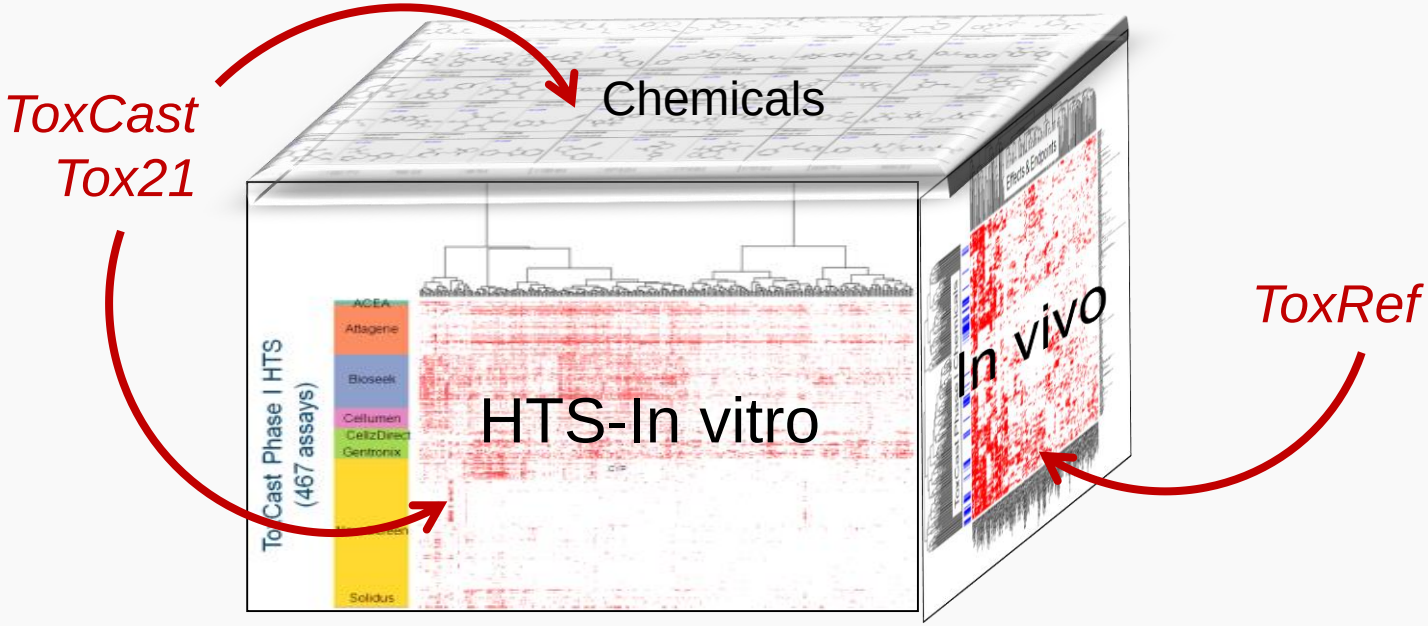
CSS Chemical Explorer Dashboard

(powered by DSSTox & ACToR)



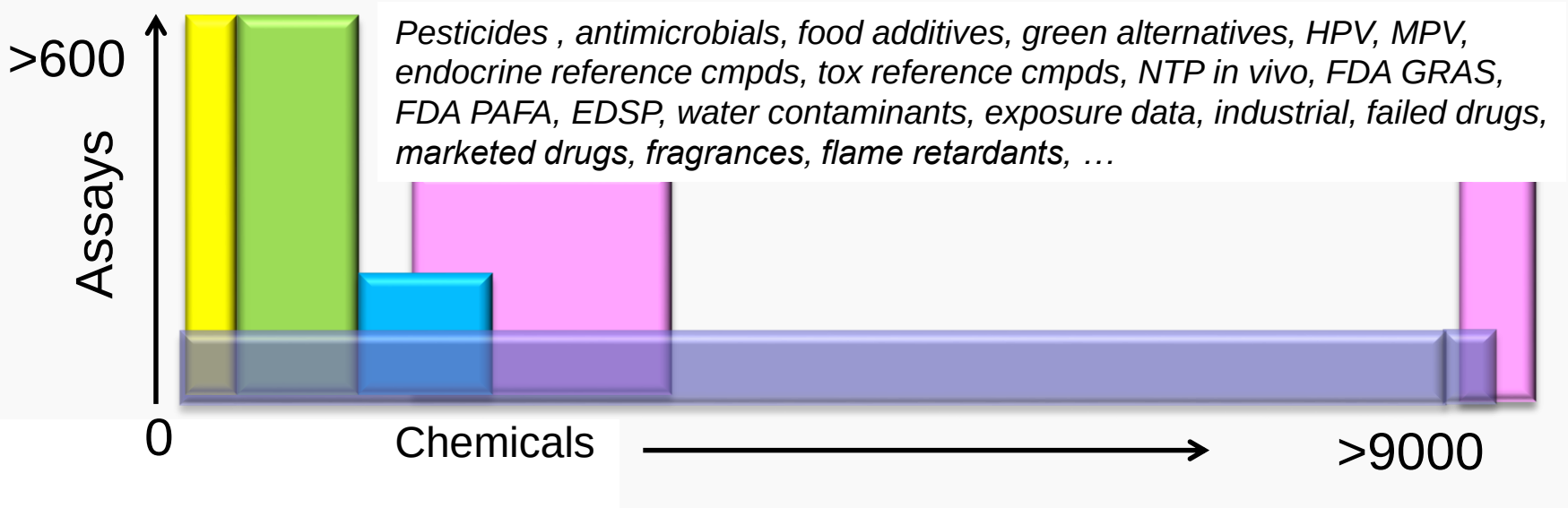
EPA's ToxCast/Tox21 Projects

- Build a diverse, highly prioritized chemical library of interest to EPA regulatory programs (e.g., EDSP) and of relevance to environmental toxicology
- Use high-throughput screening (HTS) to generate bioassay profiles and fill data gaps for thousands of chemicals
- Use all of these data to improve ability to model adverse outcomes

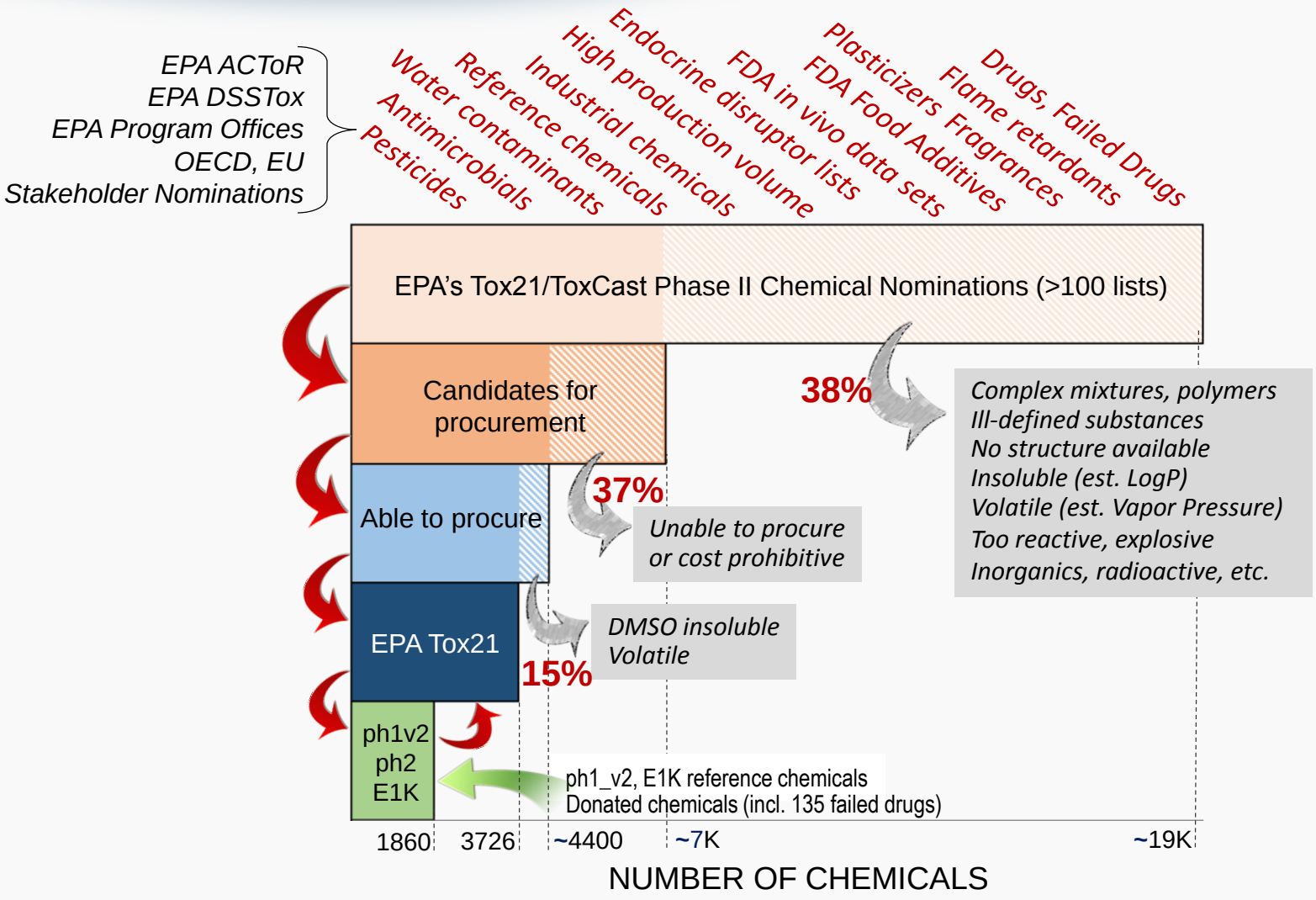


ToxCast & Tox21 Inventories

Set	Chemicals	Assays	Endpoints	Completion	Available
ToxCast Phase I	293	~600	~700	2011	Now
ToxCast Phase II	767	~600	~700	03/2013	Now
ToxCast E1K	800	~50	~120	03/2013	Now
Tox21	~8900	~80	~150	Ongoing	Ongoing
ToxCast Phase III	~2000	~300	~300	In process	2014-2015

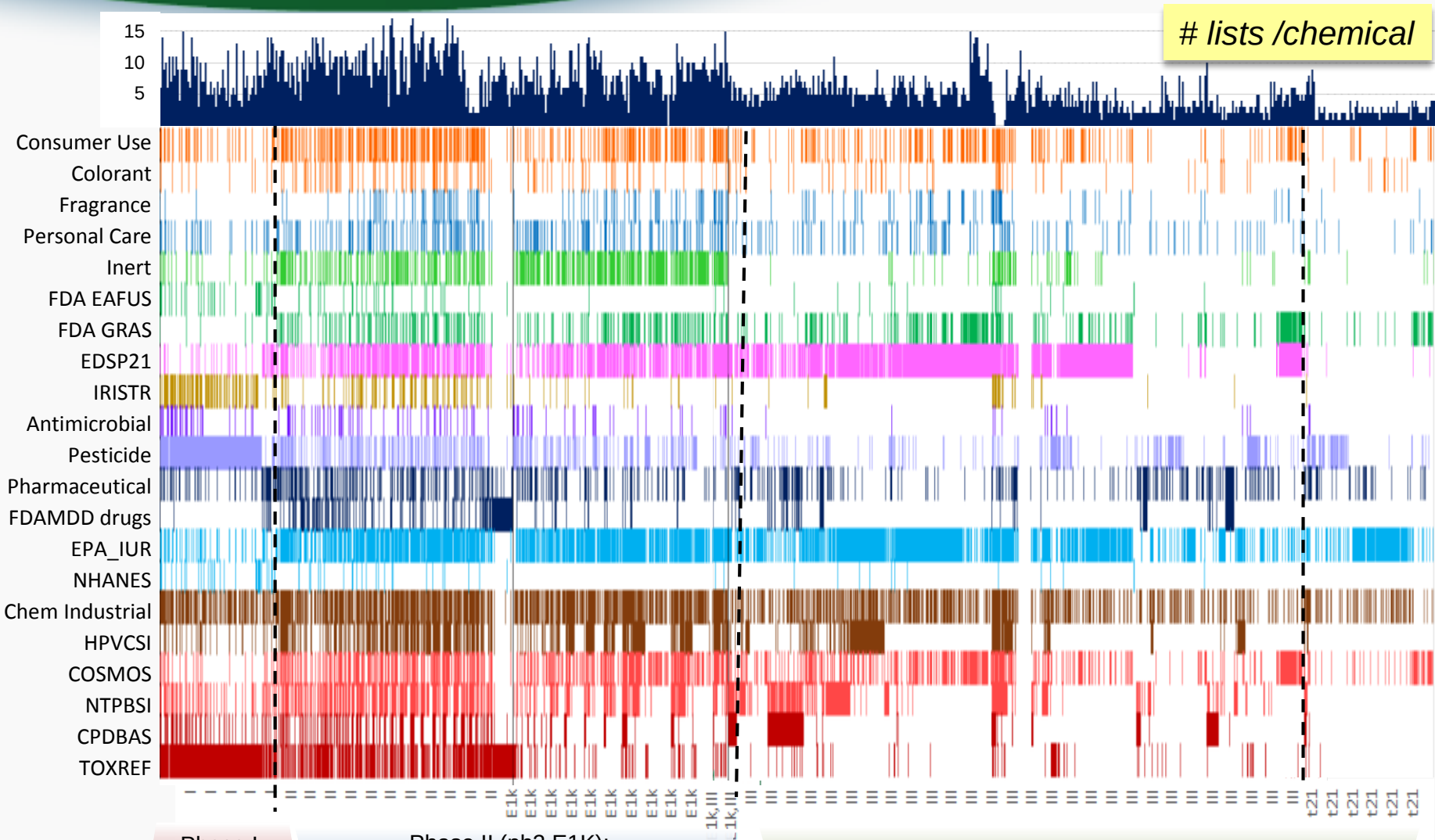


Construction of EPA's Tox21/ToxCast Inventories



ToxCast Chemical Coverage: Use, Exposure, Toxicity

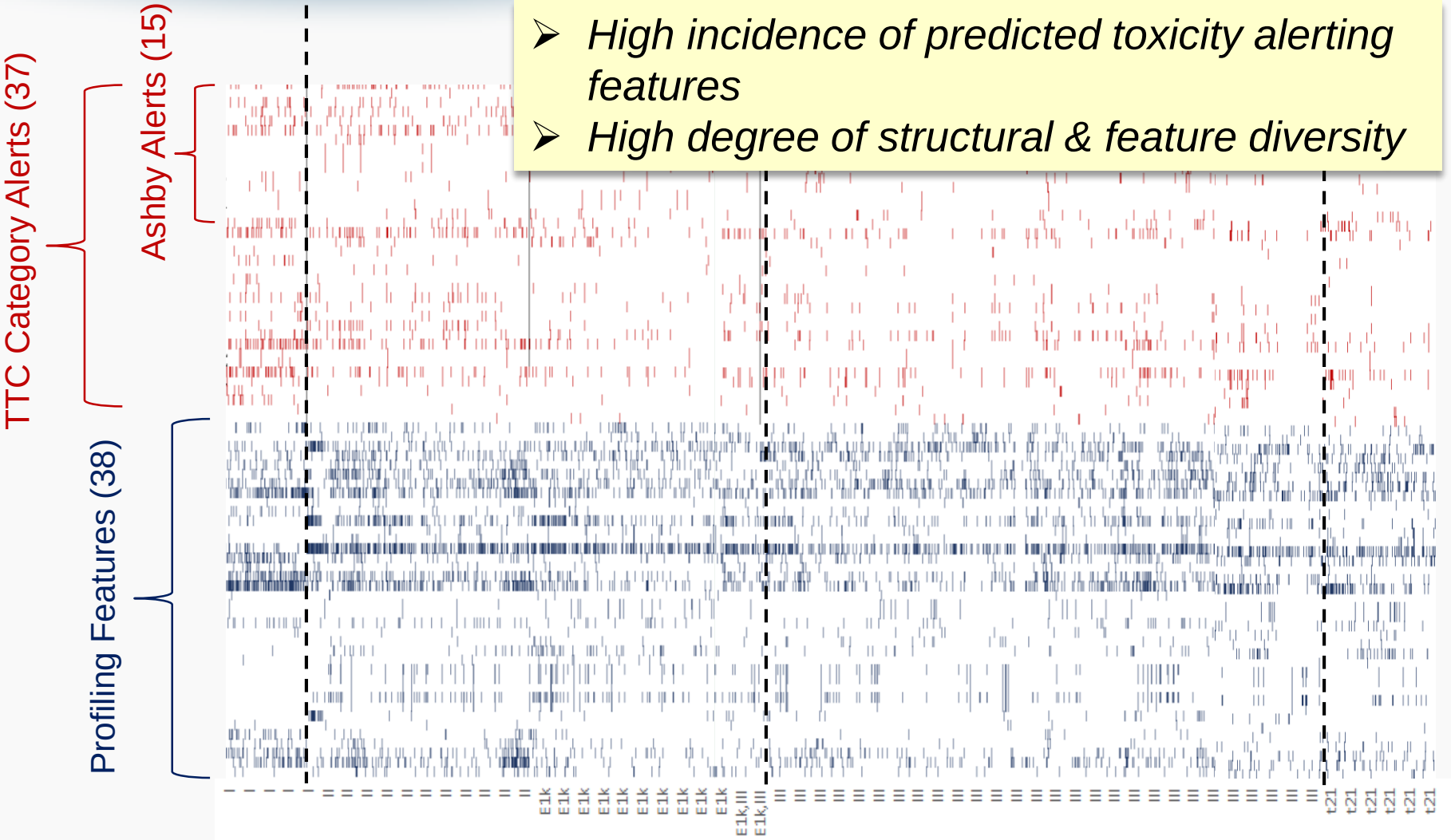
EPA United States Environmental Protection Agency



ToxCast: Toxicity Structure-Alerts & Generic Feature Coverage

EPA United States Environmental Protection Agency

- *High incidence of predicted toxicity alerting features*
- *High degree of structural & feature diversity*



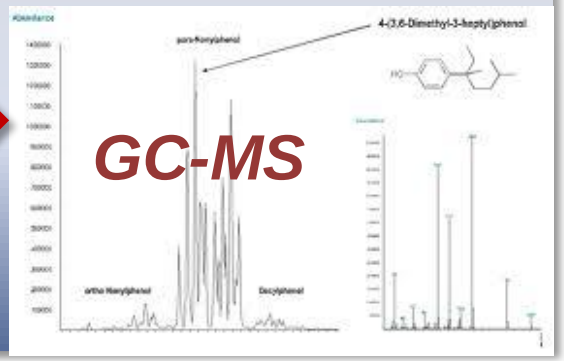
ToxCast Chemicals (4214) ordered by Testing Phase

Tox21 Analytical QC

A copy of each parent Tox21 384 well plate is subjected to analytical QC for assessing purity, identity, concentration, stability

Confirm parent ion peak and >90% purity

Fail, inconclusive or analytical method inappropriate

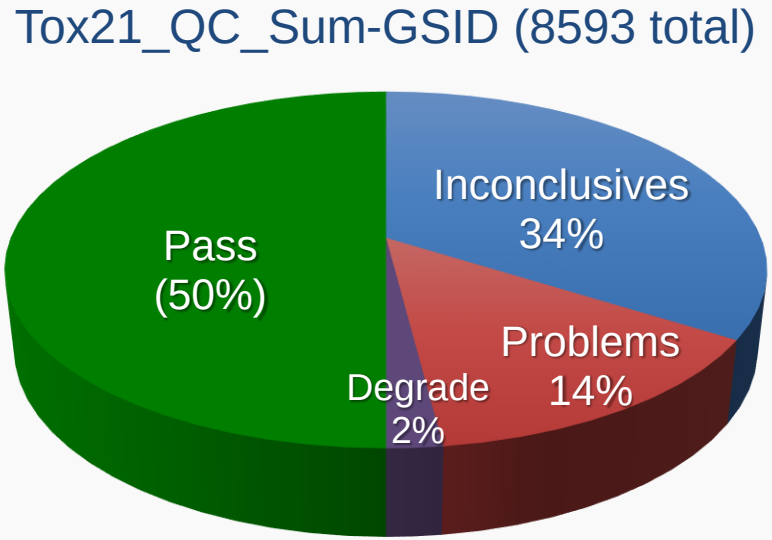


Retest at 4 mo. time point under assay conditions for stability

Publish QC summary results in association with assay data

Work performed under NIH Contract with OpAns, Durham, NC

Tox21 and ToxCast Chemical Library Analytical QC Results (8/2015)



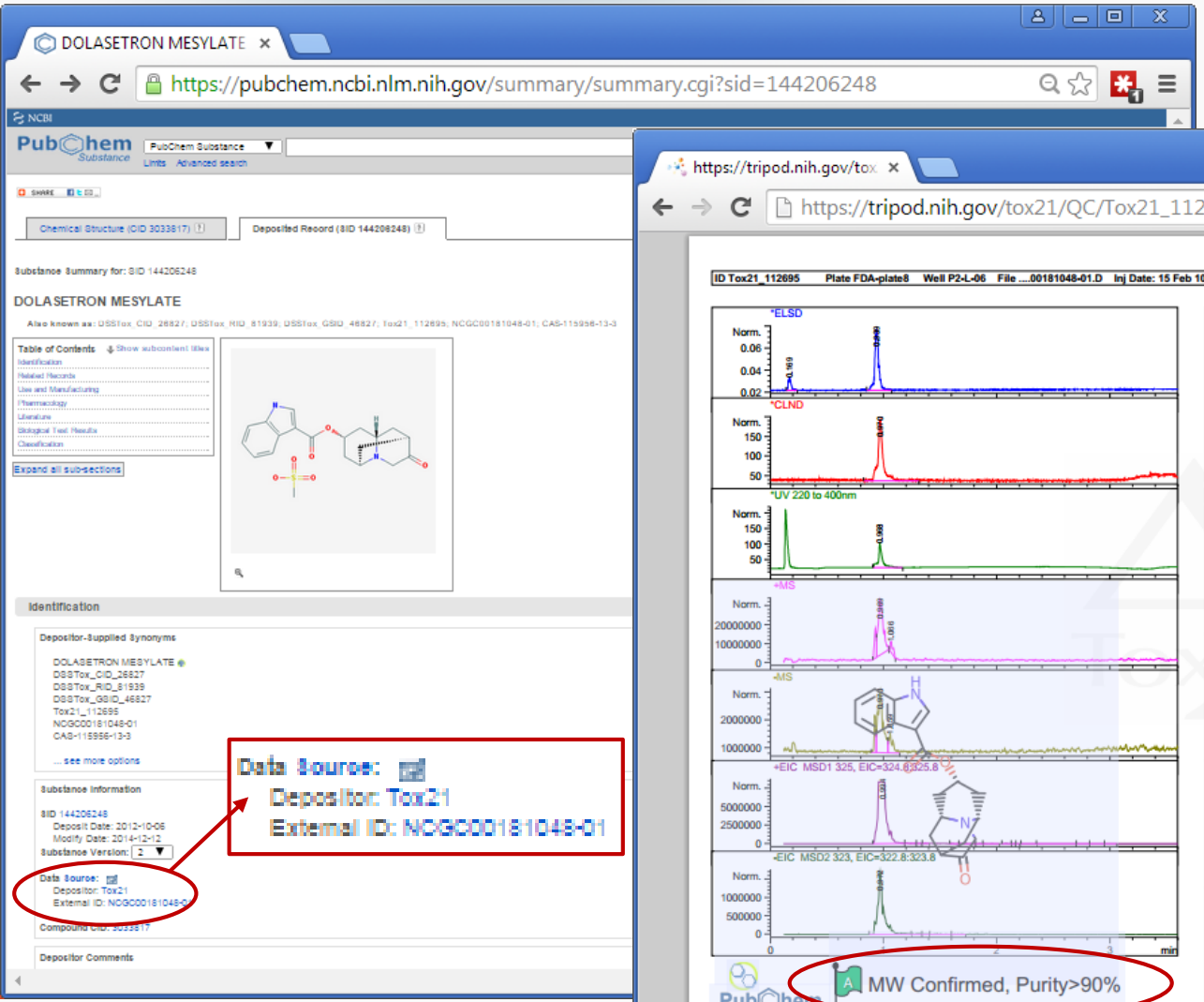
- 50% pass purity/ID/concentration checks
- A third(34%) of library pose analytical QC challenges (LC-MS and GC-MS)
- 2% degrade after 4 months under testing conditions
- 14% problems - purity (<75%), ID and/or low concentration (<30% of expected [C])

- *Which chemicals have QC issues? (e.g. SVOCs?)*
- *Which chemicals were not analyzed? (e.g., mixtures, inorganics, etc.)*
- *How are the HTS activity profiles linked to QC?*

Results as of 8/2015

Tox21 Analytical Chemical QC:
Publicly available in PubChem

EPA United States Environmental Protection Agency



Chemical “Universe” problem

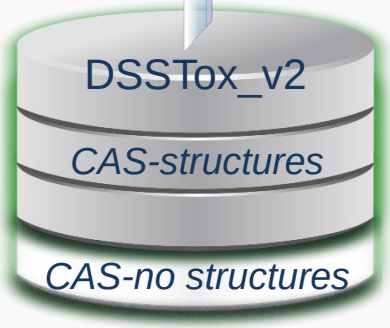
Biodegradation products
Metabolome
Protein & DNA fragments
Virtual screening libraries
Combinatorial chemistry
Polymer fragments
Polyorganic acids
Adducts, surfactants

The other 98%

Scientific literature
Toxicology studies
Environment/Industry
Commercially available

Exposure?

Toxic?



- Where should DSSTox expand chemically?
- What part of the universe should we store in databases?
- How can the valuable ToxCast physical library be shared for greatest gain?
- What cheminformatics “plumbing” would be most useful to this community?

Building EPA's Chemical Informatics Infrastructure & Linkages

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Measured Properties

Phys-chem properties

Exposure Data

Models

Reactivity Biotransformation

Fate & Transport
ADME
Toxicity Predictions
Structure-alerts

DSSTox_v2

Chemicals

NTS

water
upholstery
clothes
air
toys
dust
carpet

Biological activities

Biological Data

ACToR Inventories

CPCat, Use Exposure

ToxCast Tox21

Chemicals

HTS-In vitro

In vivo

ToxRef COSMOS

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