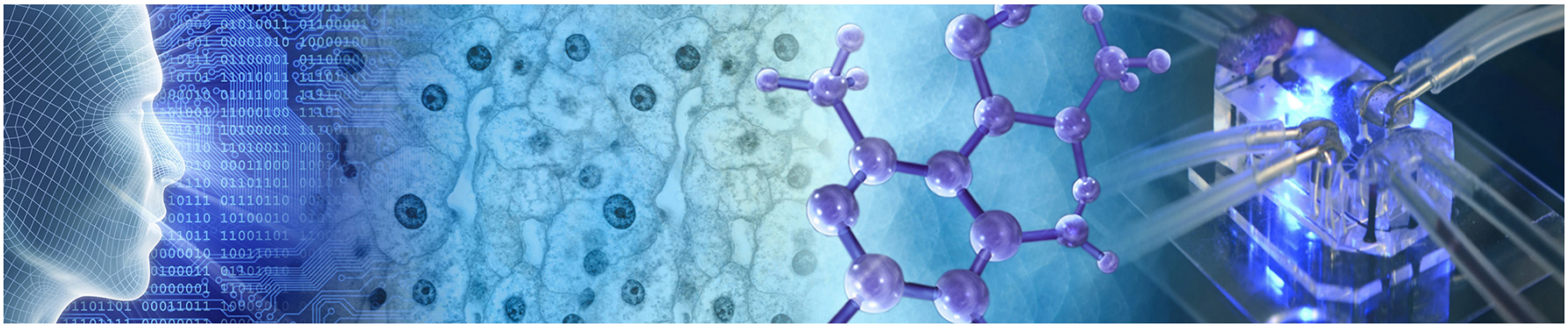
NTP
National Toxicology Program



NICEATM Update

ICCVAM Public Forum

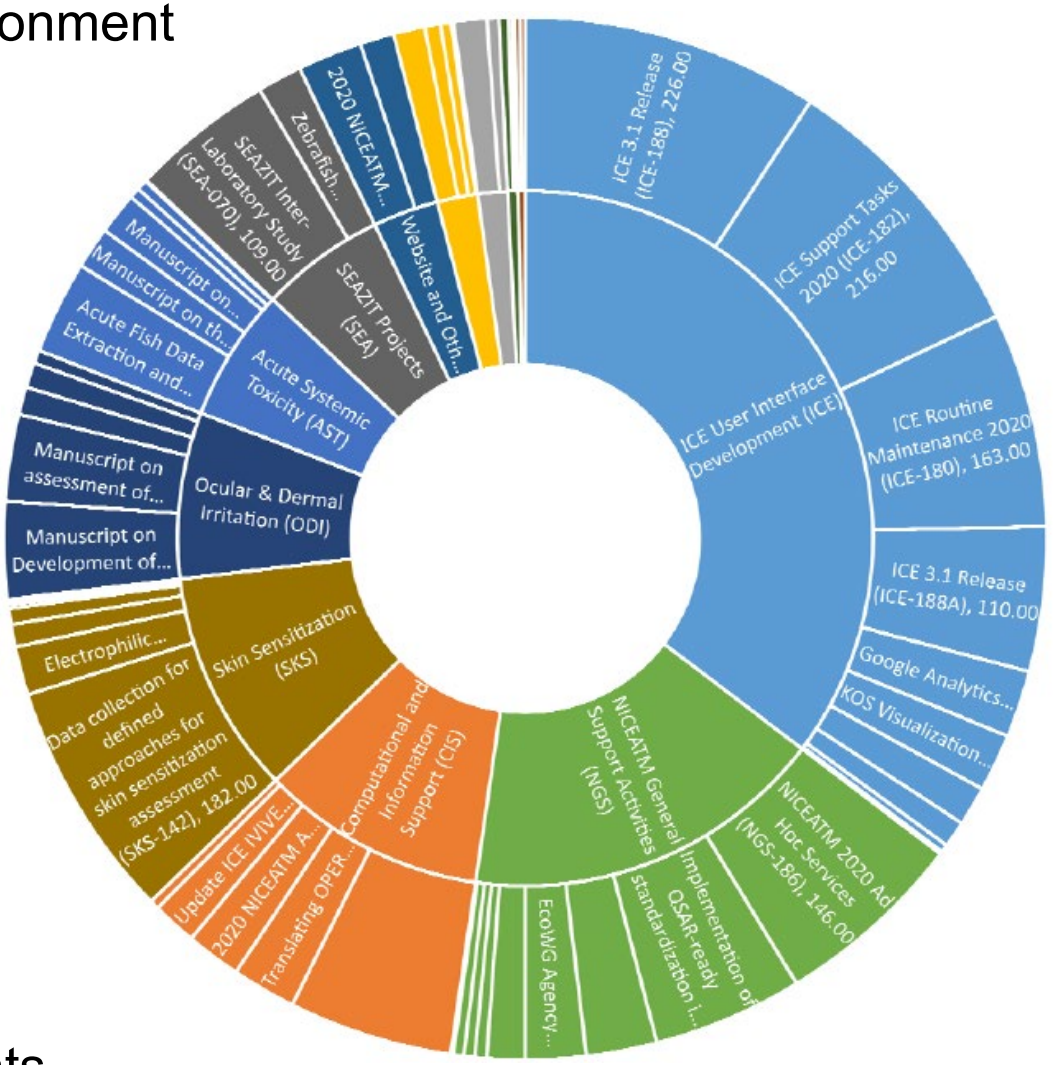
May 21, 2020

20 YEARS OF ICCVAM
Advancing Alternatives to Animal Testing

DEPARTMENT OF HEALTH & HUMAN SERVICES • USA

Ongoing Efforts

- Integrated Chemical Environment
- OPERA (QSAR/QSPR)
- Variability of in vivo data
- Data curation
- Acute Toxicity
- Dermal absorption
- Eye and skin irritation
- SEAZIT
- Skin sensitization
- Carcinogenesis
- Cardiovascular toxicity
- Animal-free affinity reagents

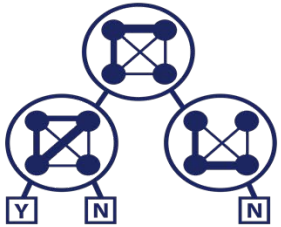


Computational Toxicology Tools

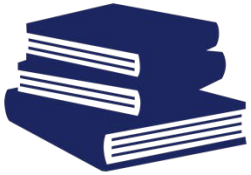
- Computational tools and resources play a critical role in chemical evaluations such as:
  - Data aggregation
  - Exploring chemical properties
  - In vitro to in vivo extrapolation
  - Mapping high-content data to biological systems
  - Generating predicted values

Integrated Chemical Environment (ICE)

Computational models



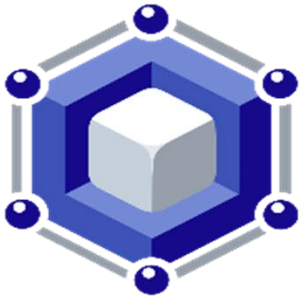
Published data



Databases

Validation studies

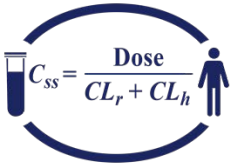
Download reference lists



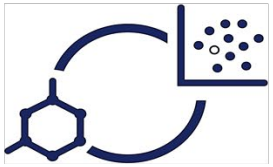
Export queries and results

Search

Data



IVIVE



Chemical characterization

https://ice.ntp.niehs.nih.gov/

Key Features/Functionality

- Data are curated and annotated
  - Provides context for those unfamiliar with a given assay
  - Removes low confidence values (e.g. due to chemical QC)
  - Provides mapping back to controlled terminology
- Tools are browser based
  - IVIVE (EPA *httk* package and in-house code) can be run with ICE data or user data from the browser, open-access, nothing to install
  - Easily merge HTS results from Tox21/ToxCast with available in vivo data
- Connection with other tools/resources:
  - Send query chemicals to EPA dashboard (bulk search July 2020) and DTXSIDs link directly to the chemical dashboard page
  - Links to CEBS under development (via API for test article information and study details, and via IVIVE tool to overlay in vivo effect levels)

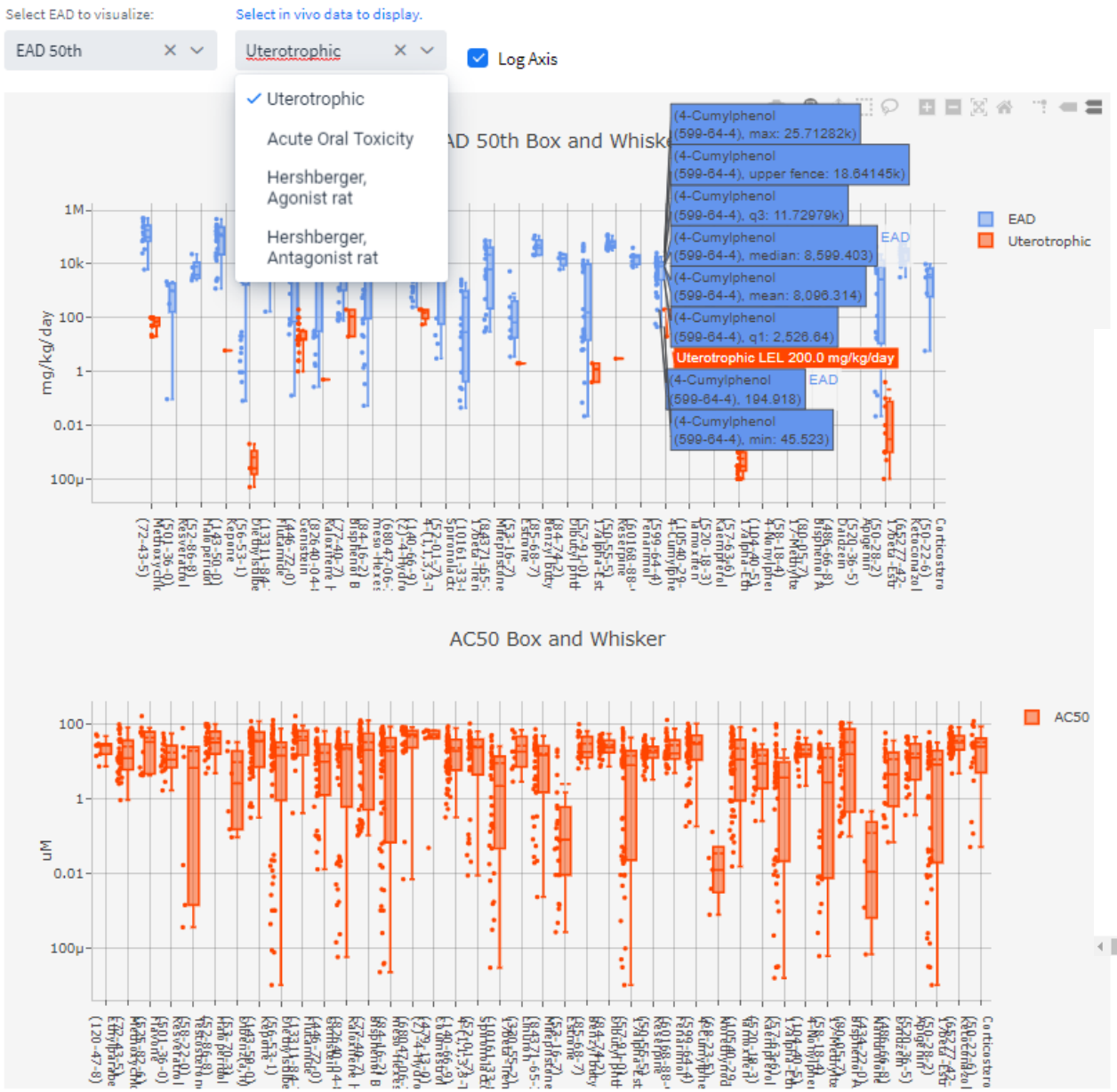
Data in ICE

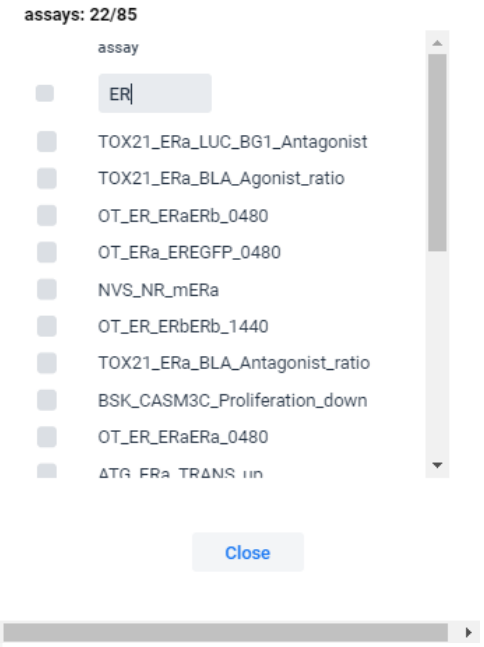
Toxicity endpoint	Assays	# of chemicals*
Acute Oral Toxicity	*In vivo* acute oral toxicity	10,335**
Skin Sensitization	DPRA, hCLAT, KeratinoSens, LLNA, human potency, etc.	578
Skin Irritation	*In vivo* acute skin irritation/corrosion, 4h HPT; *In vitro* irritation/corrosion (e.g.,EpiSkin, TER)	271
Eye Irritation	*In vivo* acute eye irritation/corrosion (e.g., Draize eye), Vitrigel	183
Endocrine	AR/ER Pathway Models, Uterotrophic, Hershberger, AR/ER transactivation	1,917**
cHTS	Curated ToxCast and Tox21 assays	9224
OPERA predictions	BP, HLC, KOA, BCF, LogP, MP, MW, VP, WS, *COMPARA*, *CERAPP, CATMoS*	838,911
Formulation data	Six-pack	298 (747 formulations)

*Values from March 2020 Release

**Does not include in silico predictions from OPERA

Overlay In Vivo Data on IVIVE Results





- Overlay in vivo data
- Update the plots with assay filtering
- Download data to explore locally

Open Structure-Activity/Property Relationship App

- OPERA is a free and open-source quantitative structure-activity relationship (QSAR) tool.
- OPERA predictions include:
  - Physchem properties
    - General structural properties
    - Environmental fate
  - ADME properties
  - Tissue partition coefficient inputs
  - Models for Toxicity Endpoints
    - CERAPP: Collaborative Estrogen Receptor Activity Prediction Project
    - CoMPARA: Collaborative Modeling Project for Androgen Receptor Activity
    - CATMoS: Collaborative Acute Toxicity Modeling Suite

https://github.com/NIEHS/OPERA

Global Collaborative Projects

Applying machine learning to predict endpoints of regulatory importance

CERAPP

Collaborative Estrogen Receptor Activity Prediction Project (2015/16)

CoMPARA

Collaborative **M**odeling **P**roject for **A**ndrogen **R**eceptor **A**ctivity **(2017/18)**

CATMoS

Collaborative **A**cute **T**oxicity **Mo**deling **S**uite **(2018/19)**

Endocrine Disruptor Screening Program (EDSP)

ICCVAM Acute Systemic Toxicity Workgroup

Mansouri et al. 2016 EHP 124:1023–1033
Mansouri et al. 2020 EHP 128 (2)
Kleinstreuer et al. 2018 Comp Tox; Mansouri et al. 2020 in prep

Agency-Based Modeling Endpoint Selection

Binary Models

Hazard

- Very 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)

Hazard

OSHA®

Collaborative Acute Toxicity Modeling Suite (CATMoS)

Original: independent calls

	VT	NT	EPA	GHS	LD50
molX	0	0	2	3	2.5

WoE: consistent calls

	VT	NT	EPA	GHS	LD50
molX	0	0	2	3	2.36

Winning bin

Model Prediction	0	5	50	300	500	2000	5000	mg/kg
VT	0	0	1	1	1	1	1	
NT	1	1	1	1	1	0	0	
EPA	0	0	1	1		0	0	
GHS	0	0	1	0	0	0	0	
LD50?	0	0	1 (160)	1	1 (613)			
WoE	**1**	**1**	**5**	**4**	**3**	**1**	**1**	

How to adjust quantitative LD50?
Avg of Lower CI and upper bin threshold

(160+300)/2
=230mg/kg

Collaboration with ATWG partners and ICCVAM agencies

Agency	No. Substances	Agency	No. Substances
Air Force	421	EPA OPP	36
Army Public Health Command	18	EPA OPPT	8
Army Edgewood Chemical Biological Center	42	EPA NCCT	4815
CPSC	110	FDA CFSAN	22
DOT	3671		

Evaluate and optimize CATMoS predictions based on lists of interest

LD50 Data Curation

Issues in acute oral tox data revealed by model predictions

C	L	M	T	V	BH	BI	BJ	BK	BL	BM	BO	BP	BQ
RML.CAS.r	Count	Original_LD50 (Concatenate)	ld50_mea	log(LD50_	ECHA_log(LD50) (Median)	CATMoS_LD50_data	CATMoS_LD50_pred	AD_LD50	AD_index	Conf_inde	Curated LD50 (mg/kg unless otherwis	New LD50(mg/kg)	ECHA dossier
106-88-7	2	>1<1.58, ca.900	635.4839	2.010766	1.53241611	2.698970004	2.853029628	1	1	0.916667	900, 1100uL/kg (so ~1100 mg/kg)	1100	https://www.e
107-83-5	1	ca.15.84			1.199755177		3.448749354	1	1	0.725	15.84, 15,840 from analog - hexane?	15840	https://www.e
109-99-9	1	1.65			0.217483944	3.217483944	3.187110886	1	1	0.953463	1.65 g/kg	1650	https://www.e
111-66-0	15	>5, >2000, >2000<5000, >5000	2841.763	0.640297			3.45444881	1	1	0.835565	5ml/kg, 10ml/kg (so ~5000mg/kg and	5600	https://www.e
111-67-1	8	>5, >5000, >5000, >5000, >500	3152.287	0.871083			3.492481795	1	1	0.829743	> 10,000 mg/kg	10000	https://www.e
111-90-0	10	<5, >5000, 5600, 6300, 6429, 7	4053.38	1.120322		3.745855195	3.65968502	1	1	0.96	6031mg/kg	6031	https://www.e
112-41-4	15	>5, >2000, >2000<5000, >5000	2841.763	0.640297			3.544496936	1	1	0.818182	> 5 600 mg/kg bw	5600	https://www.e
112-88-9	30	>5, >5, >2000, >2000, >2000<5	2792.337	0.62916			3.635710211	1	1	0.818182	>5600 mg/kg	5600	https://www.e
1120-36-1	30	>5, >5, >2000, >2000, >2000<5	2792.337	0.62916			3.596186376	1	1	0.818182	>5600 mg/kg	5600	https://www.e
120657-54	1	>5					3.666120933	1	0.939981	0.800223	>5000mg/kg based on methods sectic	5600	https://www.e
15290-77-	1	>2					2.753248503	1	1	0.928571	>2000	2500	https://www.e
15708-41-	2	ca.10, >2000	2467.803	1.798928	2.272034022	3.699056855	3.542618212	1	1	0.826087	>2000, 10000	6750	https://www.e
2082-81-7	1	1066			1.002856926	1.002856926	3.519759531	1	0.925145	0.857464	10.066 listed, but dose groups were n	10066	https://www.e
27689-12-	1	>17					3.199754313	1	0.819989	0.820274	16 mL/kg (17,600 mg/kg).	17600	https://www.e
39255-32-	3	>5, >5, >2000	2004.849	1.21517			3.706432708	1	1	0.75	>2000, >5000(MALES), >5000(FEMALE	3500	https://www.e
4499-91-6	7	>33, >300, >655, >2000, >2000	2447.428	0.74583			3.83929336	1	1	0.755952	2000, 2000, 5000, 5000, 2000, >5<15g	3500	https://www.e
543-39-5	1	5.3			0.72427587	0.72427587	3.290357289	1	0.95565	0.898544	5.3g/kg	5300	https://www.e
56-81-5	3	>20<39800, 27, 18300	11044.07	1.645202		3.958324932	3.740734556	1	1	0.68	27260 mg/kg	18300	https://www.e
592-41-6	15	>5, >2000, >2000<5000, >5000	2841.763	0.640297			3.296929233	1	0.955175	0.823902	read-across source >5600 mg/kg.	5600	https://www.e
629-73-2	30	>5, >5, >2000, >2000, >2000<5	2792.337	0.62916			3.60464617	1	1	0.818182	5ml/kg, 10ml/kg, 5g/kg, >2000<5000	3500	https://www.e
75-50-3	11	ca.2, 396.9, 397, 460, 500, 512	496.477	0.783002	2.823474229	2.662757832	2.657059529	1	1	0.806983	2.0g/kg	666	https://www.e
76114-73-	4	<2, ca.1000, >1000<2000, >=13	744.9386	1.568433	3.08804563		2.692073541	1	1	0.761905	ECHA typo lists 2mg/kg, but test dose	1250	https://www.e
7620-77-1	7	>33, >300, >655, >2000, >2000	2447.428	0.74583			3.722889223	1	1	0.794444	5g/kg, >5<15g/kg, 3g/kg, 15g/kg, 300(	3500	https://www.e
77-98-5	11	12.575, >12.5<125, 43.75, 47,	423.6832	0.5267	2.235528447		2.963016785	1	1	0.791173	>300<2000, >12.5<125, 43.75, 12.5-75	175	https://www.e
872-05-9	15	>5, >2000, >2000<5000, >5000	2841.763	0.640297			3.514069783	1	1	0.826087	5ml/kg, 10ml/kg, 5gm/kg, >2000<500(	3500	https://www.e

Examples where the 5 models (VT, NT, EPA, GHS, LD50) are in agreement with high confidence levels, with high margin between predictions and ECHA data

Variability of In Vivo Data

Rabbit skin irritation test

Prior type	I	II	III	IV
I	86.5%	4.0%	7.2%	2.4%
II	10.4%	34.9%	31.1%	23.6%
III	4.5%	4.0%	43.5%	48.0%
IV	0.6%	1.5%	9.5%	88.4%

Rabbit eye irritation test

Prior type	1	2A	2B	NC
1	73%	16.1%	0.4%	10.4%
2A	4.2%	32.9%	3.5%	59.4%
2B	0.2%	4%	15.5%	80.2%
NC	1.1%	3.5%	1.5%	93.9%

Data Curation – is the LC50 really this variable?

CASRN	LC50	LC50 unit	source
79-11-8	1268	mg/L	eChemPortal
79-11-8	0.18	mg/L	ChemIDplus

Results and discussion

Effect levels

Sex:	male/female
Dose descriptor:	LC50
Effect level:	> 1 268 mg/L air (analytical)
Based on:	test mat.
Exp. duration:	4 h

Acute Toxicity: inhalation

Currently viewing: 001 Key | Experimental result

Administrative data | Data source | Materials and methods | Results and discussion | Ap

Duration of exposure:	ca. 4 h
Concentrations:	512 (± 150) mg/m3 and 1268 (± 77) mg/m3
No. of animals per sex per dose:	5 animals per sex per dose

NIH U.S. National Library of Medicine | TOXNET TOXICOLOGY DATA NETWORK

Help | FAQs | TOXNET Fact Sheet | Training Manual & Schedule

TOXNET > ChemIDplus > Substance

Registry Number | equals | 79-11-8 | Search

ChemIDplus
A TOXNET DATABASE
Lite • Browse • Advanced

Download | Start New Query | Modify Query | Search History

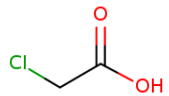
Switch to Summary View

Substance Name: Chloroacetic acid [BSI:ISO]
RN: 79-11-8
UNII: 5GD84Y125G
InChIKey: FOCAUTSVDIKZOP-UHFFFAOYSA-N

Note
Urinary metabolite of vinyl chloride.

Molecular Formula
C2-H3-Cl-O2

Molecular Weight
94.4967



3D

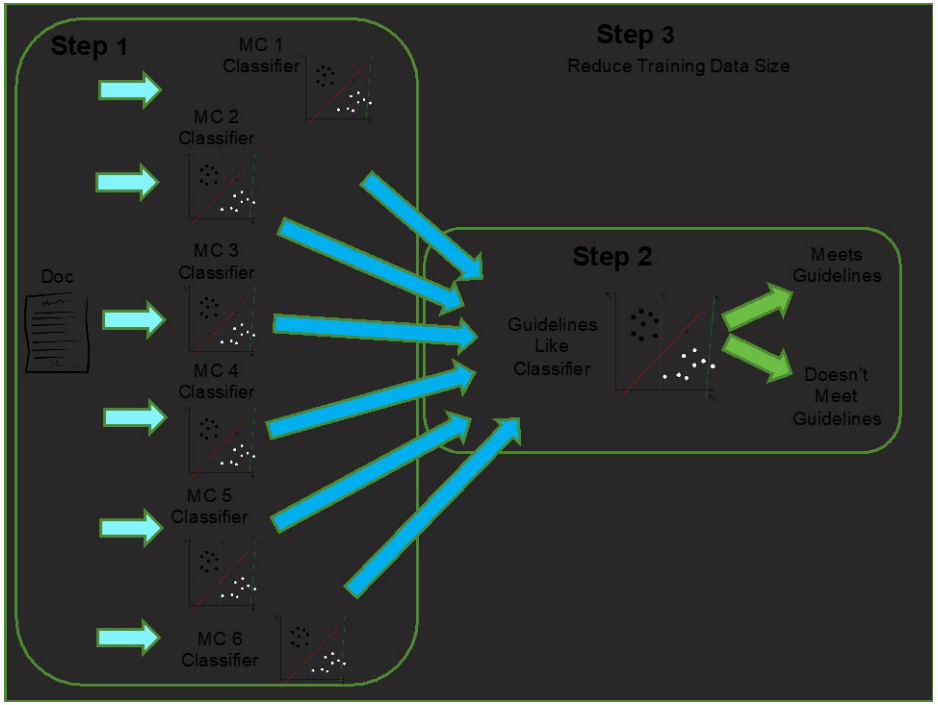
All | Classifications | Links to Resources | Names & Synonyms | Registry Numbers | Structure Descriptors | Toxicity | Physical Properties

Toxicity

Organism	Test Type	Route	Reported Dose (Normalized Dose)	Effect	Source
mouse	LD50	subcutaneous	250mg/kg (250mg/kg)		Archives Internationales de Pharmacodynamie et de Therapie. Vol. 116, Pg. 154, 1958.
rat	LC50	inhalation	180mg/m3 (180mg/m3)		Gigiena Truda i Professional'nye Zabolevaniya. Labor Hygiene and Occupational Diseases. Vol. 18(9), Pg. 32, 1974.
rat	LD50	intraperitoneal	16600ug/kg (16.6mg/kg)		Russian Pharmacology and Toxicology Vol. 41, Pg. 113, 1978.
rat	LD50	oral	55mg/kg (55mg/kg)		Gigiena Truda i Professional'nye Zabolevaniya. Labor Hygiene and Occupational Diseases. Vol. 18(9), Pg. 32, 1974.
rat	LD50	subcutaneous	5mg/kg (5mg/kg)		Toxicology and Applied Pharmacology. Vol. 22, Pg. 303, 1972.

LC50 = 1.268 and 0.18 mg/L instead?

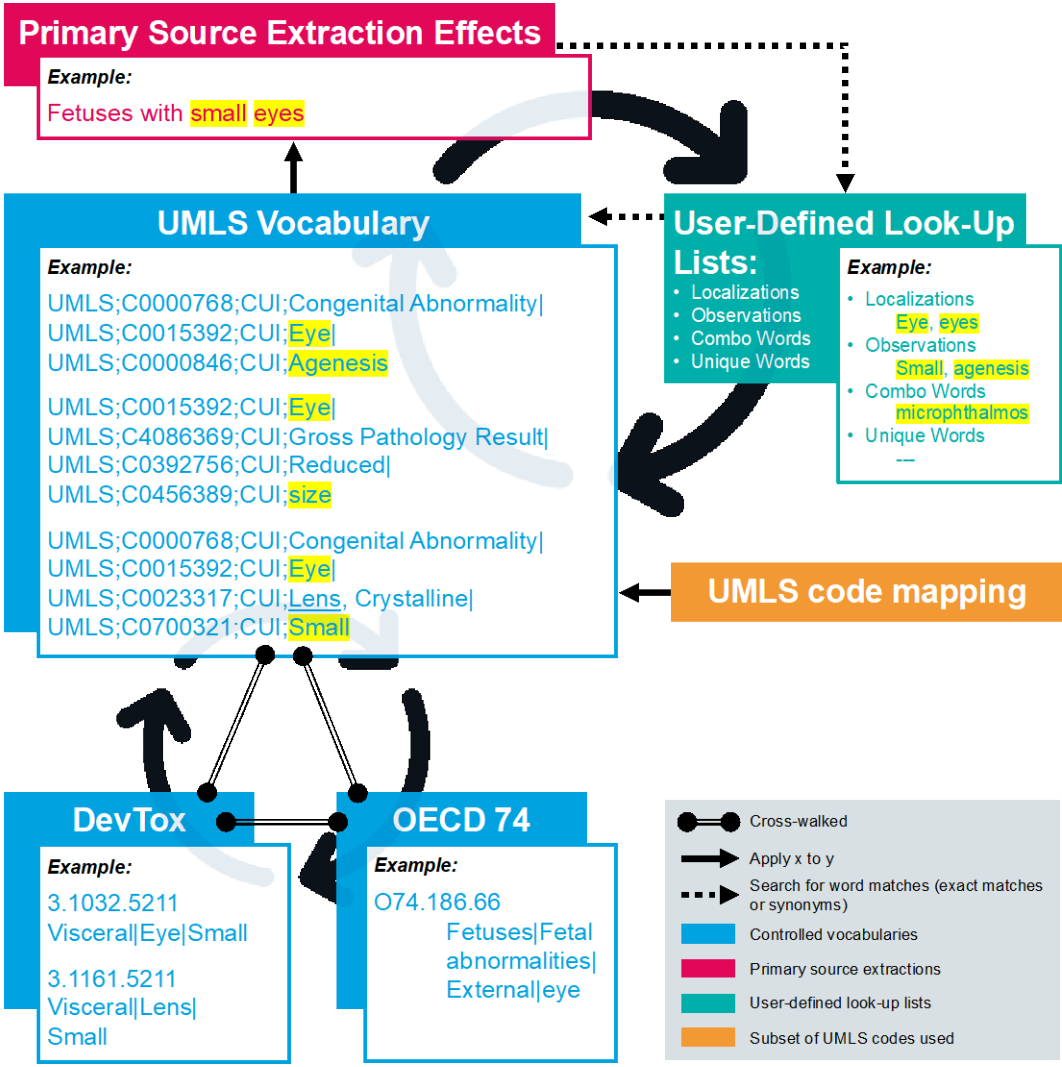
Automating Reference Data Identification



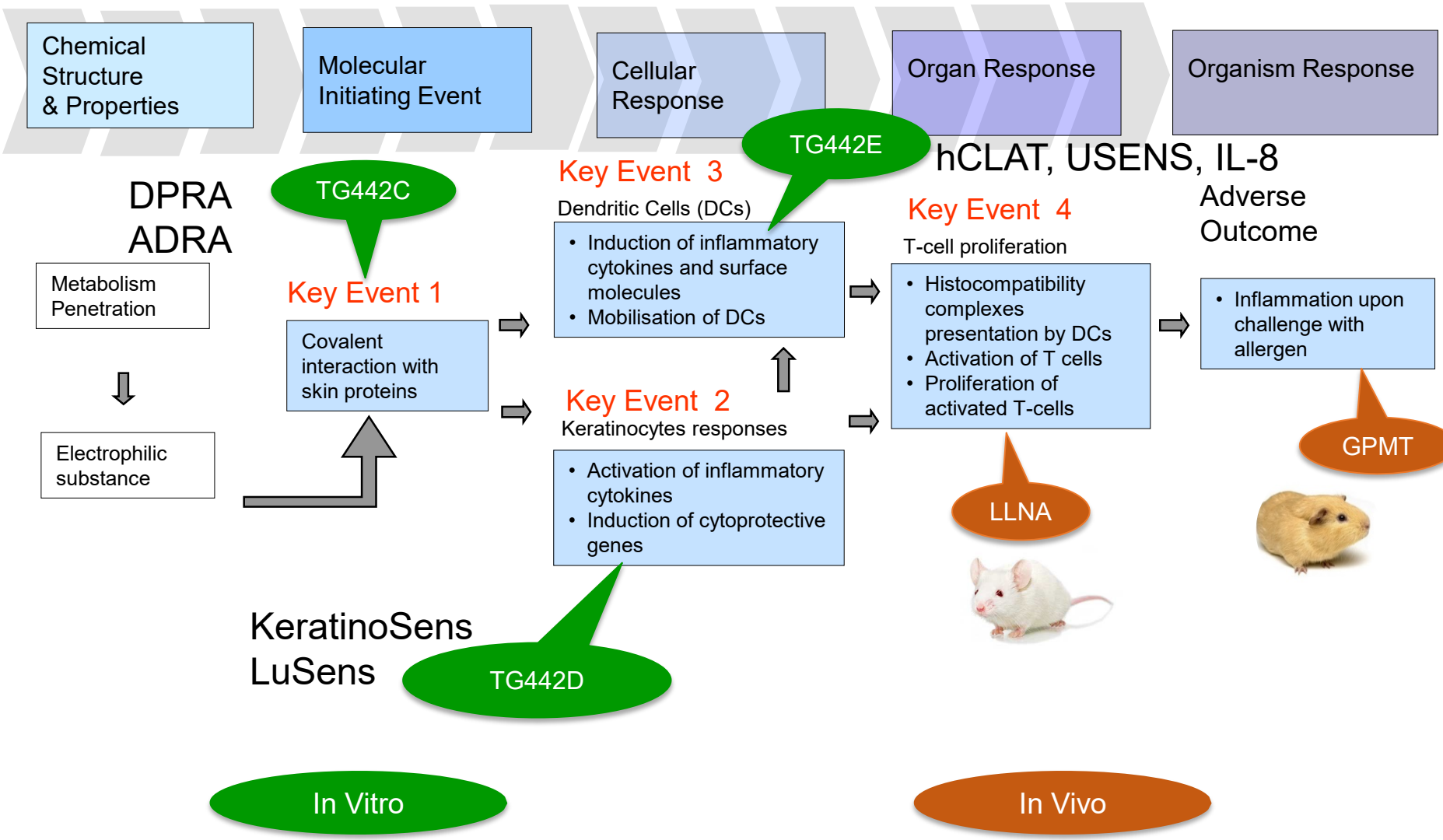
- Project with Oak Ridge National Labs (ORNL) and FDA CFSAN to apply text-mining (NLP) approaches & ML to identify high-quality data
- Semi-automated retrieval and evaluation of published literature (trained on Kleinstreuer et al. uterotrophic database)
- Apply to developmental toxicity studies (with ICCVAM DART EG)
  - Define literature search keywords, identify corpus
  - Extract/characterize study protocol details from regulatory guidelines: minimum criteria
  - Apply ML algorithms to identify high-quality studies, expert check

Study Extractions and Endpoint Mapping

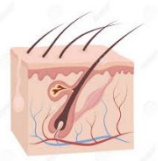
- Extract study details from prenatal developmental toxicity guideline studies
  - NTP legacy studies
  - ECHA submissions (expert reviewed for quality)
- Map results to controlled vocabularies/ontologies
  - UMLS (ToxRefDBv2.0)
  - EPA/BfR DevTox DB
  - OECD Harmonized Templates



Skin Sens Test Methods Mapped to AOP



OECD DA SS Guideline Project



- 2017 work plan: JRC/US/Canada co-leads
  - Aims to **substitute** the need for animal testing for skin sensitisation based on a combination of methods which, individually, predict key endpoint responses on the AOP: **Defined Approaches (DAs)**
  - Resulting guideline will be amenable to the agreement on Mutual Acceptance of Data (MAD)
- To meet regulatory requirements, need:
  - DAs that discriminate skin sensitisers from non sensitisers,
  - DAs that discriminate strong from moderate/weak sensitisers (GHS potency categories).
- Future work will cover DAs that address regulatory needs of quantitative risk assessment

Expert Group on DA SS

- 68 members covering regulatory authorities, OECD national coordinators, validation experts, animal welfare and industry stakeholders, method developers, etc.
- Focusing on resolution of scientific issues:
  1. **Finalization of curated reference data**
  2. **Performance comparison**
  3. **Applicability Domain**
  4. **Confidence and Uncertainty**
- Update, discussion, and feedback received at April 2020 WNT (virtual) meeting
- Virtual 2-day F2F meeting in June 2020 to discuss outstanding issues and working towards finalizing draft DA guideline
- Planning for final OECD DA guideline to be submitted for written approval by late 2020

Expanding Coverage of Chemical Space

- A significant number of chemicals used in the validation of non-animal test methods have been cosmetics ingredients
- NTP (*D. Germolec*) is supporting testing of a broad range of chemicals in internationally adopted test methods: DPRA, KeratinoSens, hCLAT.
  - Pesticide actives, agrochemical formulations, dermal excipients, personal care product ingredients, “challenge” chemicals
- Chemical nominations from multiple agencies
  - EPA: Office of Pesticides, Office of Pollution Prevention and Toxics, Office of Research and Development
  - Consumer Product Safety Commission
  - Food and Drug Administration
  - National Toxicology Program
  - Also formulations provided by Dow Chemical
- Testing began in late 2017 and will be completed in 2020

Development of a Human-Relevant Defined Approach to Assess Eye Corrosion/Irritation Potential

- Collaboration of Stakeholders EPA, NICEATM, PETA-ISC, CROs, Industry
- Reviewing available in vivo, in vitro and ex vivo test methods with respect to their relevance to human ocular anatomy, anticipated exposure scenarios, and the mechanisms of eye irritation/corrosion in humans.
- Compare/contrast to the human eye to identify features that are human relevant and to identify how they can be improved upon to increase their human relevance.
- Strengths and limitations of each method considered to assess which existing approaches are as good as or better than the currently used in vivo approach.

Ex: Damage into the corneal stroma

CELLULAR RESPONSE

Upon penetration through the epithelium and into the corneal stroma, chemicals may induce

- cell stress responses, leading to changes in cell surface markers and retraction of keratocyte cell to cell network dendrites
- necrotic or apoptotic damage
- release of chemokines and cytokines, primarily IL-1α and TNFα
- changes in relevant biomarkers
- induction of extracellular matrix / collagen synthesis
- activation of matrix metalloproteases result in loss of cell to cell adhesion and local tissue restructuring
- changes in cell metabolism/respiration
- cell death

ORGAN RESPONSE

- increased corneal susceptibility to xenobiotics
- progressive ulceration and tissue necrosis
- notable corneal swelling and swelling-related corneal opacity
- corneal opacity due to cellular/molecular denaturation/coagulation
- sloughing and loss of epithelial tissues
- induction of wound healing response and basal cell regeneration/turnover
- recruitment of neutrophils / inflammatory response
- fibrosis, pannus, and neovascularization
- loss of endothelium

ORGANISM RESPONSE

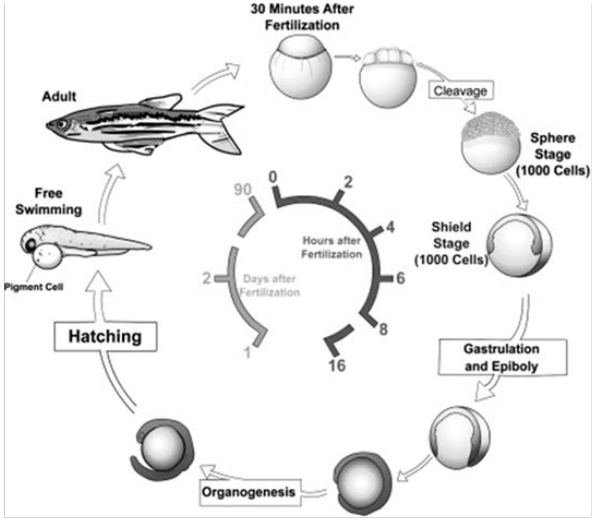
- pain and nociceptive responses
- induction of lachrymation
- permanent impact upon vision
- secondary xenobiotic exposures and biological infection
- loss of vision

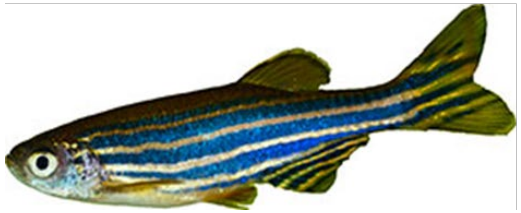
Acute Fish Toxicity – Species Comparison

- The fish acute toxicity test is used to assess potential risk to fish species, and for other ecological regulatory needs associated with surface water contaminants.
- Test is typically conducted in **three** different fish species: a cold and warm freshwater species and a marine/estuarine species.
- Can this requirement be reduced to one or two species and still provide equivalent risk protection?
- Retrospective evaluation of existing data
  - LC50 values and experimental details extracted from ~ 700 acute fish toxicity studies submitted to EPA
  - Data were analyzed to determine if there are patterns among species in term of relative differences in acute toxicity.
  - LC50 values for each chemical/species pairing assigned to EPA and UN GHS hazard categories and evaluated to determine whether the species tested influenced risk/hazard categorization.
- Results will be used to determine if reduced testing will meet risk protection goals

SEAZIT: Systematic Evaluation of the Application of Zebrafish in Toxicology

- SEAZIT's inter-laboratory study
  - 39 chemicals (3 in duplicate)
  - Dose-range finder (DRF) and definitive testing phases
- DRF data available from 3 laboratories
  - DNTP-BSB has calculated BMC's for all chemicals
- NICEATM staff analyzing DRF data
  - Exercise 1: Comparing potencies within and across labs
  - Exercise 2: Reproducibility of results within a laboratory
  - Exercise 3: Comparing results across other databases





Non-animal Derived Affinity Reagents

- Dec 2018 ESAC WG: non-animal-derived Abs can replace animal-derived Abs in the vast majority of applications.
  - EURL ECVAM report published May 15, 2020
  - Available at https://ec.europa.eu/jrc/en

- Dec 2019 NICEATM and PISC meeting: Developing strategies to increase the use of recombinant antibodies
- Coordinating with EURL ECVAM
- 4 subcommittee formed to address needed actions:
  - Education
  - Funding
  - Partnerships
  - Validation

Meeting Report

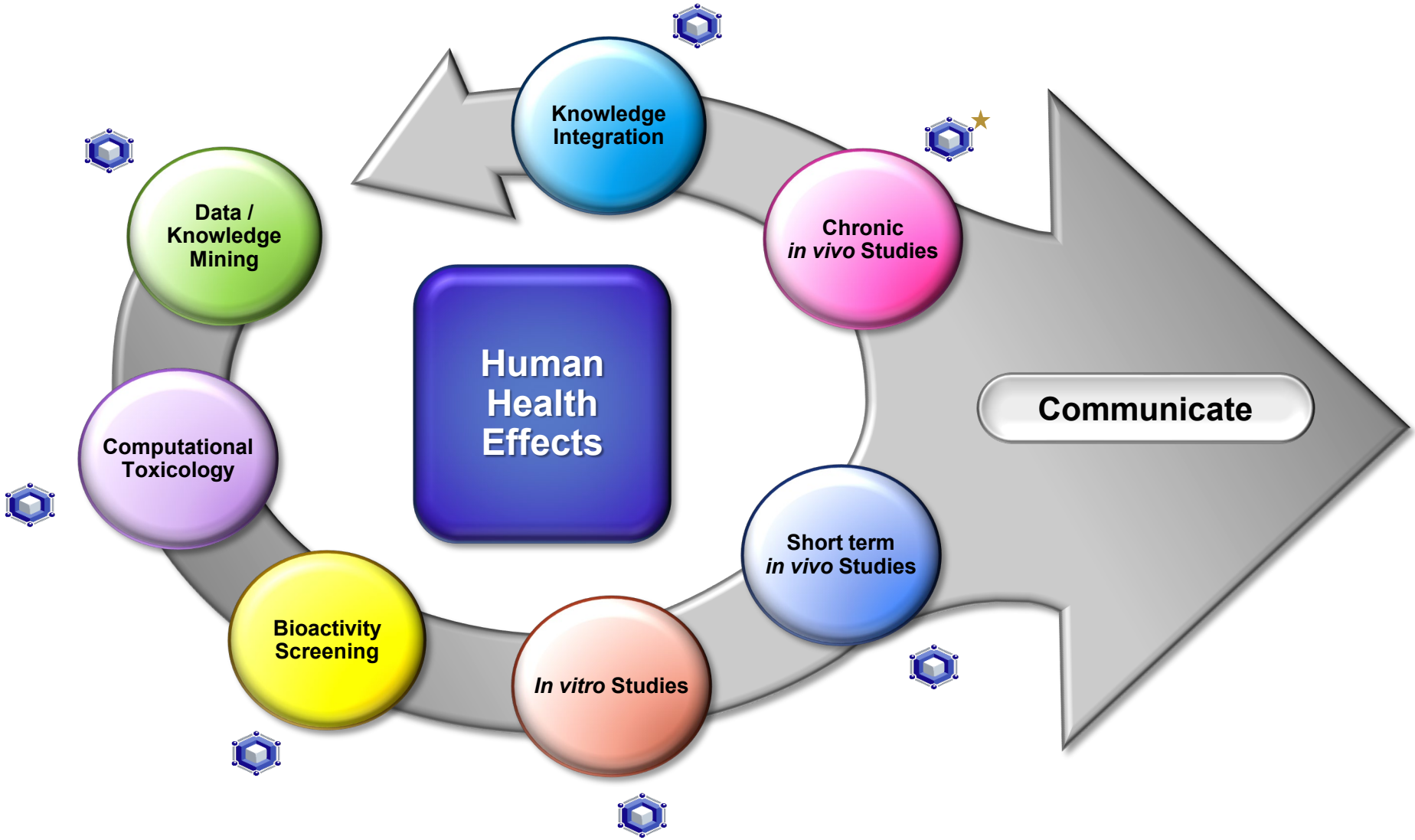
Increasing the Use of Animal-free Recombinant Antibodies

doi:10.14573/altex.2001071

Abstract

Antibodies are used in a range of research, diagnostic, and regulatory applications. Traditional methods for producing such reagents involve the immunization of animals, which introduces variability into the methods that use them and is not aligned with efforts to replace and reduce animal use. Experts from academia, biotechnology, government, and animal
e National Institutes of Health in Bethesda, MD, USA to discuss the
inant antibodies and their potential to replace antibodies derived
d the actions that resulted to facilitate increased production and use

NTP Translational Toxicology Pipeline



★ Coming Soon

Carcinogenesis HEI support activities

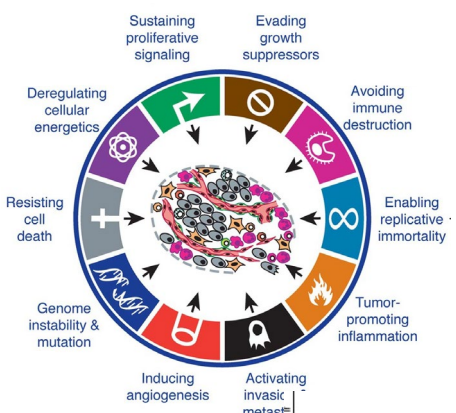
- In Silico Carcinogenicity Protocol Work Group
  - Collaborating on a position paper describing what is available/needed in terms of experimental data and computational methods for the development of an in silico carcinogenicity protocol.
  - Organization follows the Key Characteristics of Cancer (Leadscope Project, led by Ray Tice)
- Receptor Mediated Effects in Carcinogenesis Review article
  - As an extension of work done above, collaborating with Cynthia Rider (NTP) and Al Luniwal (NAMSA) to write a review article detailing the current knowledge of receptor mediated carcinogenesis.
  - Focus is collecting evidence of receptor involvement in specific cancers in human and animal models, including whether the receptor effects are true drivers of carcinogenesis (ie. A molecular initiating event or key event) or are downstream effects.
- Cancer Data Collection for ICE
  - Assembling cancer data including carcinogenicity calls from various agencies (IARC, ROC, IRIS, OPP and NTP) and specific experimental data for NTP studies extracted from CEBS and formatting for inclusion in ICE.

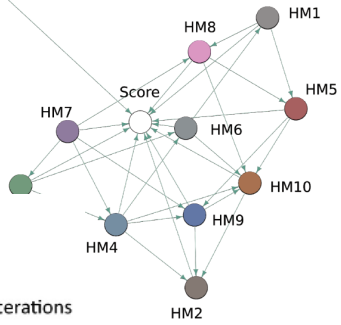
Mechanistic Mapping of HTS Assays

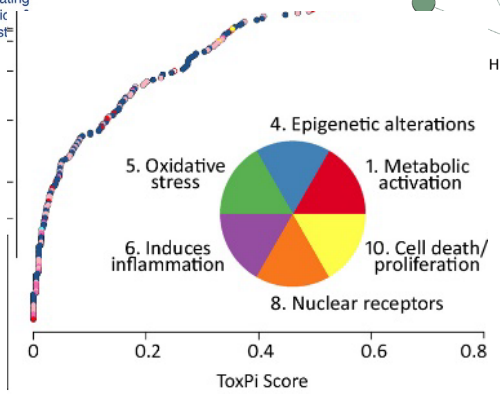
Example: Carcinogenicity

Hallmarks of Cancer & Key Characteristics of Carcinogens

- Inflammation
- Oxidative stress
- Genotoxicity/instablitiy
- Angiogenesis
- Immortalization/proliferation
- Immunosuppression
- Invasion/metastasis
- Specific receptor- or enzyme-mediated







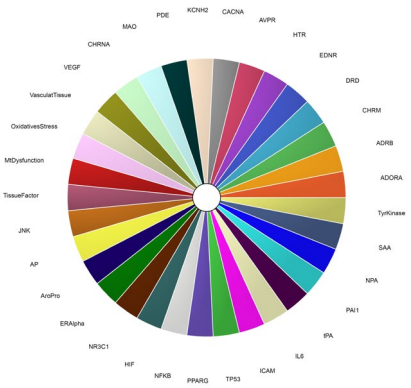
Hanahan & Weingberg 2011; Smith et al. 2016; Guyton et al. 2018; Chiu et al. 2018

Cardiovascular HEI support activities

- Identifying a pipeline of available in vitro methods targeted towards "failure modes" associated with cardiotox (*J. Santos*):
  - Changes in action potential
  - Changes in inotropy
  - Changes in vasoactivity
  - Cardiac myocardial injury
  - Valvular injury/proliferation
  - Endothelial injury/coagulation
- NICEATM to conduct literature search and data extraction
- Using Tox21 HTS data to prioritize environmental chemicals with significant activity against CV targets

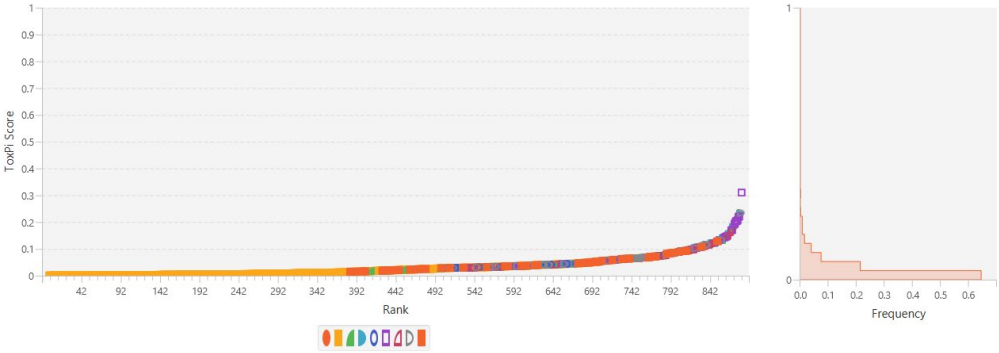
CardioToxPi: Using Tox21 qHTS Data

S. Krishna

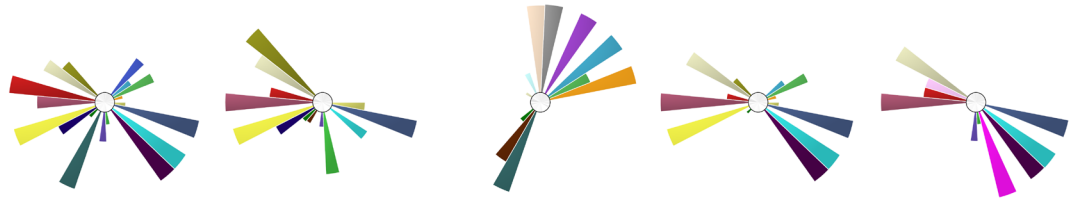


Slice	Traget name	Effect	Slice Color
ADORA	Adenosine Receptor	Vasodilation, alterations in BP	
ADRB	Adrenergic Receptor	Arrhythmia, Alterations in BP	
CHRM	Muscarinic Acetylcholine Receptor	Alterations in BP and HR, tachycardia	
DRD	Dopamine Receptor	Alterations in BP and HR, Vascular relaxation	
EDNR	Endothelin Receptor	Alterations in BP, Can exert adverse effects during	
HTR	Serotonine Receptor	Alterations in BP, Potential cardiac valvulopathy	
AVPR	Vasopressin Receptor	Alterations in BP and HR, Cardiac hypertrophy	
CHRNA	Cholinergic receptor	Alterations in BP and HR	
CACNA	Voltage-Gated Calcium Channel	Alterations in BP, QT prolongation, Arrhythmia	
KCNH2	Potassium Voltage Gated Channel	Prolongation of QT interval of ECG	
VEGF	Vascular Endothelial Growth Factor	Alterations in BP , Cardiac Ischemia	
VascularTissue	Vascular Tissue	Myocardial ischemia, cardiac Arrhythmias	
OxidativeStress	Oxidative Stress	Cellular Hypertrophy; Cardiac Cell Death	
MtDysfunction	Mitochondrial Dysfunction	Cardiac dysfunction; Cardiomyopathy	
TissueFactor	Tissue Factor	Alterations in BP and ventricular hypertrophy	
PDE	Phosphodiesterase	Alterations in cardiac contractility, HR and BP	
MAO	Monoamine Oxidase	Alterations in BP	
JNK	c-Jun N-terminal kinase	Vascular injury, cardiac hypertrophy	
TyrKinase	Tyrosine Kinase	Alterations in BP, LV dysfunction, conduction	
AroPro	Aromatase Protein	Ischemic heart disease	
ERAlpha	Estrogen receptor Alpha	Abnormal cardiac contractility, cardiac hypertrophy	
NR3C1	Glucocorticoid receptor	Alterations in BP; Arrhythmia	
	Peroxisome Proliferator		
PPARG	Activated Receptor γ	Cardiac hypertrophy , Atherosclerosis	
AP	Activating Protein	Atherosclerosis	
HIF	Hypoxia Inducible Factor 1	Ischaemia disease	
NFKB	NF Kappa B	Atherosclerosis	
TP53	Tumor Protein p53	Alteration in cardiac function	
ICAM1	Intercellular adhesion molecule 1	Markers of endothelial dysfunction	
IL6	Interleukin 6	Markers of inflammation and oxidative stress	
t-PA	Tissue Type plasminogen activator	Markers of endothelial dysfunction	
PAI -1	Plasminogen activator inhibitor type	Markers of endothelial dysfunction	
NPA	Natriuretic peptide A	Release in response to elevation in LV filling pressure	
		Direct promotion of vascular dysfunction	
SAA1	Serum amyloid A1	through SAA within vascular tissues	

Ranking of Chemicals by CardioToxPi



CardioToxPi images for 10 most active chemicals



Gentian Violet
Score = 0.3099
Rank = 882 (out of 882)

PharmaGSID_48505
Score = 0.2349
Rank = 881 (out of 882)

SSR150106
Score = 0.2313
Rank = 880 (out of 882)

Didecyldimethylammonium chloride
Score = 0.2212
Rank = 879 (out of 882)

Tributyltin chloride
Score = 0.2189
Rank = 878 (out of 882)

3-Methyl-4,6-di-tert-butylphenol
Score = 0.2079
Rank = 877 (out of 882)

SR146131 trifluoroacetate (1:1)
Score = 0.2045
Rank = 876 (out of 882)

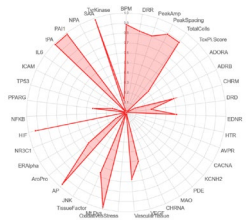
Kepone
Score = 0.2035
Rank = 875 (out of 882)

Tributyltin methacrylate
Score = 0.1994
Rank = 874 (out of 882)

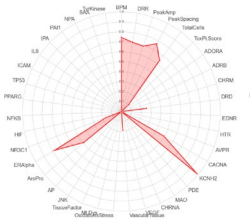
PharmaGSID_48519
Score = 0.1920
Rank = 873 (out of 882)

- CardioToxPi results were compared to PODs from iPSC-derived cardiomyocytes evaluating effects on beats per minute (BPM), decay-rise ratio (DRR), peak amplification, peak spacing, and total cell number (Sirenko et al.)
- Example chemicals are displayed below, covering those with positive effects in both HTS-based CardioToxPi and iPSC cardiomyocyte endpoints, or showing effects in only cardiomyocytes, with minimal CardioToxPi scores.

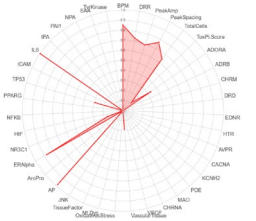
Gentian Violet



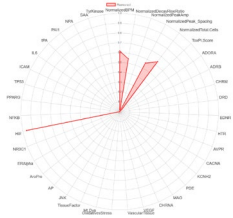
Tamoxifen



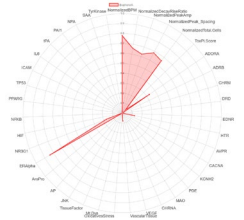
Diethylstilbestrol



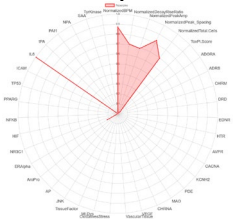
Fluorouracil



Bisphenol A



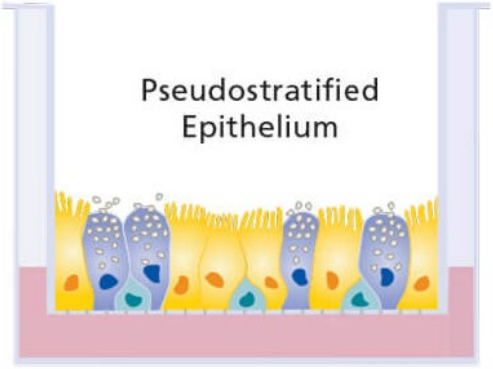
Reserpine



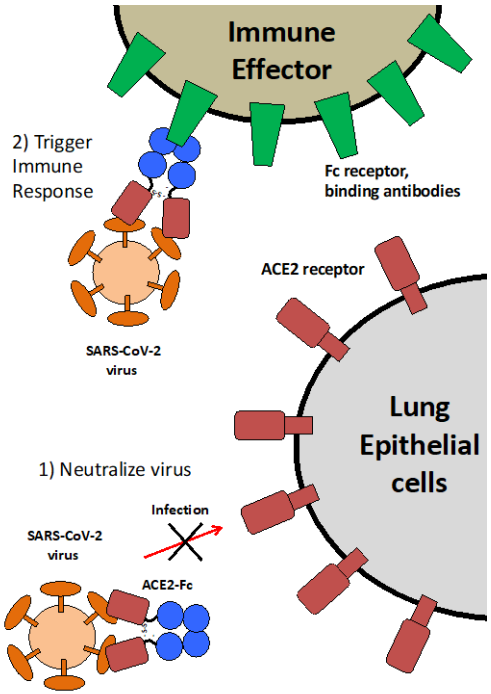
COVID-19 Therapeutic Work

Leveraging human cell-based in vitro systems

- HEK293 and MVECs
- Reconstructed Human Airway Model at Air-Liquid Interface
  - Collaboration with NIEHS/DIR (*S. Garantziotis, N. Martin*)



- Pseudostratified, ciliated epithelium that is representative of the in vivo bronchial epithelium
- Treat with soluble ACE2 receptor attached to a human immunoglobulin Fc domain as a decoy to bind the spike protein of the SARS COV-2 virus.



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- Lauren Browning

20 YEARS OF ICCVAM
Advancing Alternatives to Animal Testing