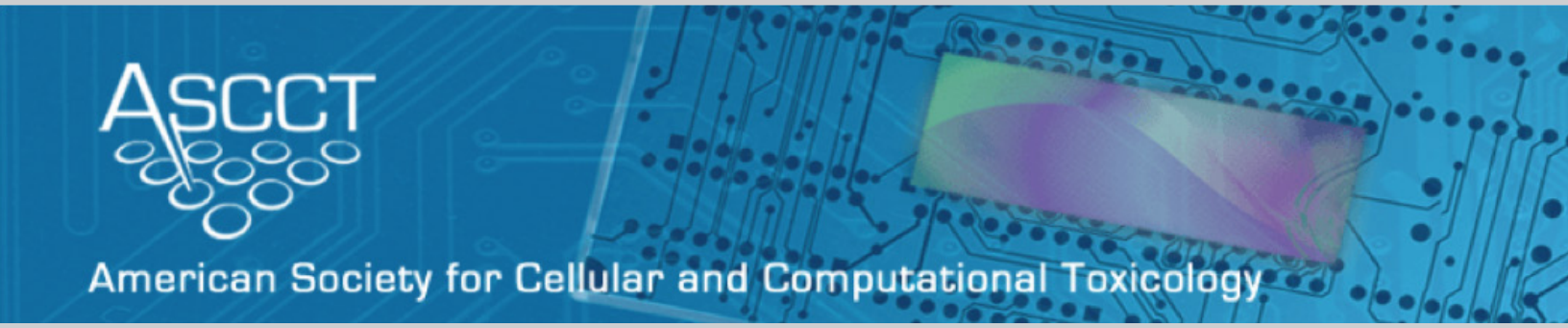

8th Annual Meeting
American Society for Cellular and Computational Toxicology

Computational Toxicology: Peeking into the Clouds while Keeping our Feet on Solid Ground

September 25th-26th, 2019

Gaithersburg Hilton - Montgomery Ballroom

Gaithersburg, Maryland

President's Welcome

Dear attendees,

On behalf of the board of directors and the organizing committee, I would like to welcome you to the 8th Annual Meeting of the ASCCT.

I recently attended the European Societies of Toxicology (EuroTox) annual meeting where a new specialty section dedicated to *in vitro* and *in silico* approaches, In2Tox, was launched. Several attendees commented on how important it is to combine these two disciplines into one specialty section as they are inter-connected. I was very pleased to hear these comments as our society was originally formed to facilitate communication and collaboration between these two areas.

The organizing committee has put together a wonderful program which has a focus on computational approaches, but will have many presentations on *in vitro* methodologies as well. New this year is a Computational Tools Showcase during which developers of these methods can share their platforms with all attendees. The meeting will conclude with a panel discussion exploring the opportunities, and challenges, of utilizing *in silico* tools. The committee has also allowed ample time to explore other topics and network during the poster sessions and reception.

I would like to thank the organizing committee and the many volunteers who helped put the meeting and mentoring event together. And a big thank you to our generous sponsors who have allowed us to keep registration fees low again this year. Lastly, but most importantly, thank you to all the speakers and attendees as your participation in the annual meeting makes the goals of the society a reality.

Welcome and enjoy the next two days!

Erin Hill
ASCCT President

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2019 ASCCT Agenda

8th Annual Meeting of the ASCCT

Computational Toxicology: Peeking into the Clouds while Keeping our Feet on Solid Ground

Tuesday, September 24, 2019

7:00 am	Pre-meeting Mentoring Event Gaithersburg Hilton, Rooks Corner Bar

Wednesday, September 25, 2019

8:30 am	Registration and Coffee
9:00 am	Welcome and Introduction *Erin Hill, President, ASCCT*
9:10 am	Protocols describing the combined use of experimental data and *in silico* results for the assessment of toxicological endpoints *Glenn Myatt, Leadscope*
10:00 am	Session I Whole transcriptome extrapolation and mechanism of action analysis: Tunicamycin case study *Ruchir Shah, Sciome* Good decisions require good data: Creating curated and structured resources for toxicology *Shannon Bell, Integrated Laboratory Systems, Inc*
10:40 am	Break
11:00 am	Characterizing the GPCR-ome target profile of emerging chemical agents with PRESTO-TANGO *Tyler DP Goralski, Combat Capabilities Development Command Chemical Biological Center* Multi-label model for toxicity prediction *Xiu Huan Yap, Wright State University* Quantitative *in vitro* to *in vivo* extrapolation using microphysiological systems: Towards predictive toxicological assessment for inhaled reduced risk products *Aditya Reddy Kolli, Phillip Morris International*

12:00 pm	**Lunch** Rooks Corner Restaurant
1:00 pm	**Poster Session**
2:00pm	**Session II** Tiered-testing strategies and the use of new approach methodologies under the amended Toxic Substances Control Act *William Irwin, U.S. Environmental Protection Agency* ReCAAP: Carcinogenicity case study waivers for food-use pesticide registration *Gina Hilton, PETA International Science Consortium* Beyond the barrier: A readily-adoptable *in vitro* model for exploring the effects of inhaled toxicants beyond the epithelium *Samantha C. Faber, University of North Carolina Chapel Hill* Development of a high-throughput iPSC-derived Liver-on-a-Chip for hepatotoxicity detection *Ryan M Reddinger, Mimetas BV*
3:30 pm	**Break / Poster Viewing**
4:00 pm	**International Collaboraion** 4:00 pm - A new society for alternatives in India *Mohammad A. Akbarsha, General Secretary, Society for Alternatives to Animal Experiments– India* 4:15 pm - Establishment of the Asian Consortium for Three Rs supported by ASCCT *Hajime Kojima, Japan Society for Alternatives to Animal Experiments* 4:30 pm - 2020 congresses: European Society for Toxicology In Vitro 2020 and WC11 *Mathieu Vinken, ESTIV President* 4:45 pm - The International Conference on QSARs (QSAR2020) *Grace Patlewicz, Environmental Protection Agnecy*
5:00 pm	**Business Meeting** - Elections, finances, other business - Cooperation with other societies
5:30 pm	Awards Ceremony & Poster Reception

2019 ASCCT Agenda

8th Annual Meeting of the ASCCT

Computational Toxicology: Peeking into the Clouds while Keeping our Feet on Solid Ground

Thursday, September 26, 2019

8:30 am	Coffee and Networking
9:00 am	The march toward big data in toxicology: Striking a balance between machine learning and Mechanistic Modeling *Nicole Kleinstreuer, National Toxicology Program Interagency Center for the Evaluation of Alternatives Toxicological Methods*
9:50 am	Session III Toxicological mechanistic inference: Generating mechanistic explanations for adverse outcomes *Ignacio Tripodi, University of Colorado, Boulder* An integrated approach to testing and assessment of chemical mixtures in the environment: The advent of Adverse Outcome Pathway footprinting *Jason C. Lambert, US EPA* The GARD platform: Cutting-edge technology using genomics and machine learning to test for various toxicological endpoints *Joshua Schmidt, Senzagen, Inc.*
10:50 am	Break
11:00 am	Computational Tools Showcase Ignacia J. Tripodi, *University of Colorado, Boulder* DAStk: Differential ATAC-seq toolkit Kamel Mansouri, *Integrated Laboratory Systems, Inc.* OPERA QSAR Models Barry Hardy, *Edelweiss Connect* SaferSkin David Filipovic, *Michigan State University* PyPK for PBPK modeling

	Tom Luechtefeld, *Insilica* Sysrev.com Nancy Baker, *Environmental Protection Agency* Abstract Sifter Adam Lee, *Dupont* Chemical Awareness Toolkit Shannon Bell, *ILS/NICEATM* Integrated Chemical Environment (ICE) Maggie Coombs, Lhasa Limited Multiple T.J. Bozada, ToxTrack Chemchart Entrerprise
12:00 pm	Panel Discussion: Data and methodology quality and transparency Moderators: David Allen, Integrated Laboratory Systems, Inc. Panelists: Nicole Kleinstreuer, NICEATM Glen Myatt, Leadscope, Inc. Thomas Hartung, Johns Hopkins University/CAAT Kristie Sullivan, Physicians Committee for Responsible Medicine David Filipovic, MSU
1:00 pm	ASCCT Meeting Ends
1:00-5:30 pm	POST-MEETING HANDS-ON TRAINING IN COMPUTATIONAL METHODS Offered by the PCRM at Gaithersburg Hilton Separate registration requested The training will feature short summaries of key tool features followed by rotating demos with opportunities to try out the models and/or hands-on exercises. Agenda and registration for this session can be found at www.ascctox.org/meetings. Lunch will be served.

Thank you to the meeting sponsors!

ASCCT

American Society for Cellular and Computational Toxicology

Mission:

The ASCCT is a scientific society which provides an organized forum for discussion of cellular and computational toxicology approaches, especially as replacements for animal-based toxicology methods. Through its meetings and activities, the Society facilitates the development, acceptance, and routine use of cellular and computational methods through open dialog between industry, academic, advocacy, and regulatory scientists. The Society strives to include the participation of young scientists to promote their contributions to the field.

Goals:

Facilitate the development, acceptance, and routine use of cellular and computational methods.
Increase the routine application and use of computational and *in vitro* methods for prioritization, classification, and risk assessment purposes.

Foster open dialog between industry, academic, advocacy, and regulatory scientists throughout North America.
Include the participation of young scientists to promote their contributions to the field.
Strengthen cooperation between stakeholders.

All Members will receive:

A regular e-newsletter
Access to a growing library of educational webinars from field leaders
Discounted subscription rates to the journals ALTEX and *Toxicology In Vitro*
Discounted registration for ASCCT events
News and event updates in the *in vitro* and computational toxicology fields
The chance to network with regulators, scientists, and policymakers on the cutting edge of non-animal toxicology

www.ascctox.org info@ascctox.org

Table of Contents

ASCCT 2019
Invited Speakers

Protocols Describing the Combined Use of Experimental Data and *In Silico* Results for the Assessment of Toxicological Endpoints

Glenn Myatt

In silico toxicology is an important alternative approach to animal testing that provides a fast and inexpensive prediction of toxicity. While computational approaches can quickly calculate a prediction, the process of selecting and acquiring models, performing an expert review, integrating the information, and documenting conclusions and uncertainties can be time-consuming and difficult to repeat. Combining the *in silico* results with experimental data is commonly needed and this process is not well defined. It is also challenging to defend the results, primarily due to a lack of published procedures for performing an *in silico* assessment. To support the development of such protocols, a 60-member international cross-industry consortium has been assembled that includes representatives from international regulatory agencies and government research laboratories in the United States, Canada, Japan and Europe, as well as large companies from various industrial sectors (e.g., pharmaceutical, food, cosmetics, agrochemicals), academic groups and other stakeholders. The protocols will ensure any *in silico* assessments are performed in a consistent, repeatable, well-documented and defendable manner to support their broader acceptance. This presentation will outline the *in silico* toxicology protocol framework and illustrate the progress of the consortium in generating specific protocols for a number of major toxicological endpoints such as genetic toxicity and skin sensitization.

Leadscope, Inc.

The March Toward Big Data in Toxicology: Striking a Balance Between Machine Learning and Mechanistic Modeling

Nicole Kleinstreuer

Advances in computational tools now provide unprecedented access to collecting, curating, and analyzing large datasets, enabling machine learning models that deliver predictions across the chemical universe. Such machine learning approaches are often contrasted against building mechanistic models based on biological knowledge. This talk will examine the application of machine learning methods to address problems in predictive toxicology, from identifying high-quality reference data to global crowdsourcing approaches to model adverse health outcomes, as well as the construction of mechanistically informative tools and models. The argument will be presented that effective computational toxicology approaches should leverage both machine- and mechanism-based strategies. Multiple examples pertaining to acute and chronic toxicities will highlight the way that computational toxicology is changing the environmental health landscape, not only through how new models are being built, but how data is being collected, curated, and combined.

National Toxicology Program Interagency Center for the Evaluation of Alternatives Toxicological Methods

ASCCT 2019
Oral Abstracts

Whole Transcriptome Extrapolation and Mechanism of Action Analysis: Tunicamycin Case Study

Ruchir Shah[1], Deepak Mav[1], Dhiral Phadke[1], Michele Balik-Meisner[1], Mihir Shah[1], Austin Ross[1], Jason Phillips[1], Bob van de Water[2], Scott Auerbach[3], B. Alex Merrick[3], Richard S. Paules[3]
ruchir.shah@sciome.com

Targeted transcriptomic analyses decrease the time and financial burden associated with obtaining whole transcriptomic data in high throughput screens. The TempO-Seq S1500+ platform(s), now available for human, mouse, rat and zebrafish, measures a few genes that are representative of biological and pathway co-regulation across the entire genome in a given species. While measurement of these genes alone provides an overall assessment of gene expression activity, extrapolating expression values to the whole transcriptome (~26 K genes in humans) has the ability to estimate measurements of non-measured genes of interest and increases the power of pathway analysis algorithms by using a larger background genetic space. Here we use a human S1500+ Tunicamycin dataset with ~3K representative genes as a case study to explore the improved downstream analysis achieved by extrapolating expression to the remaining transcriptome. Extrapolation increased the number of significant genes by 49%, bringing to the forefront many genes and pathways that have been established to be differentially expressed by Tunicamycin exposure without significantly changing the sample variability. We demonstrate that gene- and pathway-level biological interpretations were improved by extrapolating from the ~3K measured genes to approximately 26K genes before performing a variety of downstream applications, including differential expression analysis, gene set enrichment pathway analysis, DAVID keyword analysis and Ingenuity pathway analysis. The extrapolated data highlight the role of metabolism/metabolic pathways, the endoplasmic reticulum, and the unfolded protein response, each of which are key activities associated with Tunicamycin exposure that were underrepresented in the original dataset. Therefore, our case study suggests an approach to extend and enhance data from the S1500+ platform for improved insight into biological mechanisms and functional outcomes of diseases, drugs, and other perturbations.

[1]Sciome LLC, RTP, NC, [2]Leiden Academic Centre for Drug Research, Leiden University, Leiden, The Netherlands, [3]National Institute of Environmental Health Sciences, RTP, NC

Good Decisions Require Good Data: Creating Curated and Structured Resources for Toxicology

Shannon Bell[1], Neepa Choksi[1], Patricia Ceger[1], Amber Daniel[1], Agnes Karmaus[1], Jaleh Abedini[2], David Allen[1], Warren Casey[2], Nicole Kleinstreuer[2]
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The world is more data-driven than ever, with real-time data streams used to optimize everything from music selection and GPS routes to watering and pesticide application in agriculture. Data are also used to evaluate existing or new test substances for possible harm and risk. Modern toxicity testing couples existing data with new assay results using computational methods and models to aid in decision making. This integration paradigm is used to characterize interactions between test substance and target organism to generate an effect prediction. Trustworthiness and appropriateness of data play a key role in impacting the reliability of the predictions. Less reliable data may be acceptable for prioritization or screening purposes, but when predictions are to be used as the basis of a regulatory decision, data reliability and appropriateness for the decision are critical. The NTP Interagency Center for the Evaluation of Alternative Test Methods (NICEATM) is establishing reliable curated datasets for test method developers and risk assessors. In this presentation we provide examples of how adding data structure through controlled vocabularies and expert groupings can make data accessible and guide appropriate use, particularly for users without in-depth knowledge of test systems. Additional examples highlight the importance of curation for data interpretation. Access to curated NICEATM data via the Integrated Chemical Environment is demonstrated in case studies. This work was funded in whole or in part with federal funds from the NIEHS, NIH under Contract No. HHSN273201500010C.

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Characterizing the GPCR-ome Target Profile of Emerging Chemical Agents with PRESTO-TANGO

Tyler DP Goralski, Jennifer R Horsmon, Kyle P Glover
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G protein-coupled receptors (GPCRs) function as important intermediaries of cellular signaling and have been identified as targets for a number of different therapeutics. Likewise, the function of some GPCRs are directly or indirectly affected by nerve agents and pharmaceutically based agents (e.g. opioids). Recently, a high-throughput *in vitro* system called PRESTO-TANGO was developed to interrogate the activity of an emerging threat agent against the entire human GPCR-ome. As a result, researchers now have the ability to discover novel GPCR targets with a proposed or known function, as well as agonists for orphan GPCRs. We validated a "miniaturized" 384-well high throughput format of the full PRESTO-TANGO GPCR library against various opioid and adrenergic compounds. The results generated from cells expressing opioid and adrenergic GPCRs treated with known agonists serve as proof of principle that PRESTO-TANGO is an acceptable tool for measuring and comparing efficacy between agonists of the same receptor. Ultimately, this *in vitro* system will serve as a predictive toxicological tool, allowing us to gain a full picture as to which GPCRs are inhibited or activated in cells treated with various chemical weapons, and it may provide further insight into mechanisms of action before any testing needs to be performed *in vivo*.

Combat Capabilities Development Command Chemical Biological Center

Multi-label Model for Toxicity Prediction

Xiu Huan Yap, Michael Raymer
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Most computational predictive models are specifically trained for a single toxicity endpoint. Since more than 1300 toxicity assays have been reported in the TOXCAST dashboard, achieving high coverage over this growing number of toxicity endpoints remains challenging. Furthermore, single-endpoint models lack the ability to learn dependencies between endpoints, such as those targeting similar biological pathways, which may be used to boost model performance. In this study, we characterize the performance of 3 multi-label classification (MLC) models, namely Classifier Chains (CC), Label Powersets (LP) and Stacking (SBR), on Tox21 challenge data. These MLC models employ the Problem Transformation approach, which is algorithm-independent and thus generally compatible with existing classifiers. Using Logistic Regression as the base classifier and random label partitioning (k=3), CC and LP show statistically significant improvement in model performance using Hamming and subset 0/1 scores ($p<0.05$). On the other hand, SBR show significant improvements in micro-averaged F1 and accuracy scores. Additionally, when using label partitioning learned by Louvain community detection method, CC and SBR models retain their performance improvement over single-endpoint models, suggesting that learning marginal dependence is a key contributor to improving model performance. Taken together, MLC models could potentially boost the performance of current single-endpoint predictive models. Future studies include examining the relationships among endpoints that lead to optimal performance for particular MLC methods, testing these models on datasets with a greater number of toxicity endpoints, and using base classifiers that are well-established in predictive toxicology.

Wright State University

Quantitative *In Vitro* to *In Vivo* Extrapolation Using Microphysiological Systems: Towards Predictive Toxicological Assessment for Inhaled Reduced Risk Products

Aditya Reddy Kolli, Florian Martin, Julia Hoeng
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Early prediction of *in vivo* toxicological outcomes from *in vitro* results is of enormous interest as the toxicological assessment paradigm begins to incorporate readouts from microphysiological systems (MPS). MPS are rapidly developing *in vitro* systems that mimic and allow exploration of complex *in vivo* biological responses to external stimuli. The data from multiple sources needs to be cohesively analyzed and requires adaptation of quantitative *in vitro* to *in vivo* extrapolation (QIVIVE) methodologies. We will present the existing methods and challenges involved in applying QIVIVE methods utilizing MPS that could potentially be applied for toxicological assessment of inhaled reduced risk products. The development of a QIVIVE methodology requires integration of multiple models and techniques such as quantitative structure activity relationship (QSAR), quantitative adverse outcome pathway (qAOP), particle deposition and physiological based toxicokinetics modeling. Some of the the challenges associated in performing QIVIVE are (1) *in vitro* to *in vivo* metabolism and clearance estimates; (2) *in vitro* chemical-chemical interaction analysis, (3) accounting for population variability; (4) impact of metabolite accumulation in MPS; (5) qAOP event thresholds; (6) determining deposited dose of an inhaled chemical; (8) protein expression, binding affinities and mechanistic description of cellular pathways (9) the reliability and acceptability of MPS by regulatory bodies. The application of QIVIVE methodologies for reduced risk products assessment may offer novel mechanism for risk assessment in the context of tobacco harm reduction.

PMI R&D, Philip Morris Products S.A., Neuchatel, Switzerland

Tiered-testing Strategies and the Use of New Approach Methodologies Under the Amended Toxic Substances Control Act

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On June 22nd, 2016, the Frank R. Lautenberg Chemical Safety for the 21st Century Act amended the Toxic Substances Control Act (amended TSCA). The amendments included specific provisions that mandate the U.S. Environmental Protection Agency (EPA) to encourage and facilitate "the use of scientifically valid test methods and strategies that reduce or replace the use of vertebrate animals while providing information of equivalent or better scientific quality and relevance that will support regulatory decisions." Under section 5 of the amended TSCA, EPA receives pre-manufacture notices (PMNs) on new chemical substances that have diverse chemistries, uses, and hazard concerns. PMNs are often submitted with limited toxicology data; however, EPA has developed and continues to develop chemical categories with specific structural alerts to inform the hazard potential of data poor chemical substances. This presentation will provide an overview of new chemical categories under development for chemistries that have potential hazard concerns for effects on the lungs, including surfactants, insoluble polymers, polycationic substances, and lung waterproofing agents. These chemical categories are characterized by common physical-chemical properties across diverse chemical spaces/boundaries. For example, surfactants have the common chemical property of lowering the surface tension of water, lung overload substances have the common property of insolubility in water, cationic substances have a high positive charge density, and waterproofing agents repel water. This presentation will also provide examples of how tiered testing with New Approach Methodologies (NAMs) such as *in silico, in chemico, in vitro*, and targeted *in vivo* studies can be utilized to regulate substances with lung effects under the amended TSCA. These NAMs follow the principles of the three R's (Reduction, Refinement, and Replacement) to implement vertebrate animal abatement strategies under the amended TSCA as described in the Strategic Plan to Promote the Development and Implementation of Alternative Test Methods Within the TSCA Program (Link).

Disclaimer: The views of the authors of this abstract are those of the authors alone and do not represent Agency policy or endorsement. Mention of trade names of commercial products should not be interpreted as an endorsement by the U.S. Environmental Protection Agency.

United States Environmental Protection Agency

ReCAAP – Carcinogenicity Case Study Waivers for Food-Use Pesticide Registration

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Reviews of the rodent cancer bioassay over the past 40 years have raised questions about its relevance and regulatory need to assess risk to human health. As a result, a working group comprised of experts from government, industry, and non-government organizations have formed the Rethinking Carcinogenicity Assessment for Agrochemicals Project (ReCAAP) to evaluate the appropriateness of waiving rodent bioassays for the registration of food-use pesticides. This presentation will provide case study examples of carcinogenicity waivers formulated through a weight of evidence-based (WoE) approach. The WoE includes information on pesticide exposure, mode-of-action, physiochemical properties, and sub-chronic toxicological data from defined endpoints. The results of these WoE reviews will be used to establish the criteria for when the mouse and/or rat cancer bioassay can be waived with sufficient confidence to protect public health.

Beyond the Barrier: A Readily-Adoptable *In Vitro* Model for Exploring the Effects of Inhaled Toxicants Beyond the Epithelium

Samantha C. Faber[1], Nicole A. McNabb[2], Pablo Ariel[3], Shaun D. McCullough[4]
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While epithelial cells play a well-defined role in the effects of inhaled chemical exposures, exploring toxicological effects on cell types that reside beneath the epithelium (i.e., stromal cells) and play critical roles in lung structure/function has typically required the use of *in vivo* animal models. The development of organ-on-a-chip systems has offered the potential to reduce the use of animal models; however, cost, complex engineering, and small sample size limit their availability to most researchers and applicability to experimental methods that are commonly used to inform adverse outcome pathway (AOP) development. To address this, we developed a novel *in vitro* approach to study "trans-epithelial" exposures in a readily-available, scalable, and cost-effective model. We hypothesized that trans-epithelial chemical exposure mediates oxidative stress/pro-inflammatory response within fibroblasts through an imbalance in redox signaling and alterations in cellular stress-responsive pathways. Using gene expression techniques, multi-color fluorescent live-cell imaging, and inhibitor studies we identified molecules governing key cellular signaling pathways (i.e., Nrf2 and MAPK) and defined the kinetic profile of oxidative stress/pro-inflammatory response within both epithelial and fibroblast cells. Further, spatio-temporal analysis identified a role for stromal cells in mediating pro-inflammatory signaling and redox imbalance in adjacent epithelial cells, as well as underlying molecular mechanisms. This model system is the first to enable characterization of the dynamics of oxidative stress/pro-inflammation following trans-epithelial chemical exposure, providing novel insight for the development of therapeutic interventions to reduce adverse effects of inhaled chemical exposure and provide accurate *in vitro* to *in vivo* extrapolation for inhalation toxicity testing.

[1]Curriculum in Toxicology and Environmental Medicine, University of North Carolina at Chapel Hill, [2]Department of Environmental Toxicology, University of California, Davis, [3]Microscope Services Laboratory, Department of Pathology and Laboratory Medicine, University of North Carolina at Chapel Hill, [4]Clinical Research Branch, USEPA/NHEERL

Development of a High-Throughput iPSC-derived Liver-on-a-Chip for Hepatotoxicity Detection

Ryan M. Reddinger[1], Richard DeBiasio[2], Mark Miedel[2], Lawrence Vernetti[2], D. Lansing Taylor[2], Paul Vulto[1], Albert Gough[2], Anthony D. Saleh[1] and Kristin M. Bircsak[1]
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Hepatic injury accounts for two-thirds of drug development failures in the pharmaceutical industry. The current regulatory-accepted models for assessing hepatotoxicity include rodent models which are expensive, low-throughput and overall have unreliable concordance with human hepatotoxicity, as well as standard two-dimensional (2D) *in vitro* systems (liver cancer cell lines and primary human hepatocytes), which have only marginally improved predictivity. This study sought to optimize a 3D *in vitro* model of the human liver and predictive hepatoxicity assays by adapting the vascularized Liver Acinus MicroPhysiology System (vLAMPS) developed by the University of Pittsburgh Drug Discovery Institute into MIMETAS' high-throughput organ-on-a-chip platform. Extracellular matrix (ECM) gels can be contained in the microchambers of the OrganoPlate® by capillary pressure barriers (PhaseGuides™), allowing for solid tissue and barrier formation, as well as perfused tubular endothelial vessel structures to be grown in the medium perfusion channel. The resulting platform contains 96 x 3D microfluidic co-culture biomimetics of the liver sinusoid including FujiFilm Cellular Dynamics International (CDI) induced pluripotent stem cell-derived hepatocytes (iPSC hepatocytes; iCell Hepatocytes 2.0) and stellate cells incorporated in an ECM protein gel, fed by microfluidic nutrient perfusion from an adjacent endothelial and Kupffer cell-lined blood vessel mimic. Viability of the co-culture remained stable (80-95%) for up to 21 days of culture. We report long-term maintenance of metabolic activity including CYP3A4, as well as albumin and urea production (1-21 days, both up to 20 µg/day/106 cells). Further, we report a significant drop in AFP production over the 21-day culture, as a key indicator of iPSC hepatocyte maturation in the OrganoPlate®, that is not observed in standard 2D and 3D culture of these cells. Using multi-parameter high-content imaging, we have optimized toxicity assays with fluorescent readouts to measure viability, mitochondrial function, and steatosis following the administration of known hepatotoxicants, including acetaminophen (APAP). The cell viability assay revealed up to an 80% reduction in the viability of APAP treated OrganoPlate® cultures while 2D cultures were unaffected by the same concentration of the hepatotoxicant. These studies display the feasibility of using our iPSC-derived 3D human liver model as a high-throughput screening platform for the assessment of pharmaceutical and environmental hepatotoxicity.

[1]Mimetas BV, the Netherlands, [2]University of Pittsburgh Drug Discovery Institute, US

Toxicological Mechanistic Inference: Generating Mechanistic Explanations for Adverse Outcomes

Ignacio J. Tripodi[1], Tiffany J. Callahan[2], Robin D. Dowell[3], Lawrence E. Hunter[4]
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Government regulators and others concerned about toxic chemicals in the environment hold that a mechanistic, causal explanation of toxicity is strongly preferred over a statistical or machine learning-based prediction by itself. We thus present a mechanistic inference engine, which can generate hypotheses of the most likely mechanisms of toxicity, using gene expression time series on human tissue and a semantically-interconnected knowledge representation graph. We seek enrichment in our manually-curated list of high-level mechanisms of toxicity (e.g. "Triggering of caspase-mediated apoptosis via release of cytochrome C", or "Mitochondria-mediated toxicity by inhibition of electron transport chain"), represented as causally-linked ontology concepts. Our knowledge representation consists of an integration of concepts from multiple ontologies (GO, PRO, HPO, ChEBI, PATO, DOID, CL), as well as relevant concepts from Reactome, the cellular toxicogenomics database (CTD), and the AOP Wiki. The expression assays were obtained from the Open TG-Gates, EU-funded CarcinoGenomics projects, and other relevant public datasets consisting of human liver, lung, nasal, buccal, bronchial, and kidney cells exposed to a sizeable number of chemicals that elicit different mechanisms of toxicity. Both our knowledge graph and experimental transcriptomics data are human-centric. Besides predicting the most likely mechanisms at play from the transcriptomics assays, we generate mechanistic narratives that link the most significant genes at each time point, to each of the steps in the most likely mechanisms. This provides a transparent, putative explanation of the mechanisms of toxicity, that would help inform a researcher's decision-making and aid further experimental design.

[1]University of Colorado, Boulder. Computer Science / Interdisciplinary Quantitative Biology., [2]University of Colorado, Denver. Computational Bioscience., [3]University of Colorado, Boulder. Molecular, Cellular, and Developmental Biology / Computer Science., [4]University of Colorado, Denver. Computational Bioscience / Computational Pharmacology.

An Integrated Approach to Testing and Assessment of Chemical Mixtures in the Environment: The Advent of Adverse Outcome Pathway Footprinting

Jason C. Lambert
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Human health assessment of environmental chemical mixtures has perpetually been limited by the availability of adverse health outcome data to inform hazard and dose-response assessment. That is, traditional mixtures assessment theory and practice, such as dose additivity, places emphasis on similar toxic mode or mechanism of action however as this information is rarely available for most environmental chemicals, the comparison of toxicological similarity among mixture component chemicals is typically driven to the level of phenotypic effect or apical health outcome or endpoint. The virtual absence of whole mixtures data at environmentally relevant exposure levels or proportions, and, limited availability of traditional toxicity bioassay information for individual mixture component chemicals, has resulted in very few mixtures assessments that support some type of regulatory decision-making. With the advent of New Approach Methods toxicity testing and data, such as adverse outcome pathway (AOP), opportunities to evaluate hazards associated with exposure to 'data-poor' mixture chemicals have advanced significantly. Potential health impact(s) of mixture chemicals may be informed using an integrated read-across approach that includes AOP 'footprinting' when adverse health outcome data derived from traditional bioassays are lacking. In brief, AOP footprinting makes use of key event data to inform qualitative and quantitative relationships between mixture components. This presentation will provide an overview of AOP footprinting and how it compliments an integrated approach to testing and assessment (IATA) of environmental chemical mixtures.

The views expressed in this abstract are those of the author and do not necessarily reflect the views or policies of the U.S. Environmental Protection Agency.

U.S. EPA

The GARD™ Platform–Cutting-Edge Technology Using Genomics and Machine Learning to Test for Various Toxicological Endpoints

Joshua Schmidt[1], Andy Forreryd[2], Henrik Johansson[2]
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Predictive toxicology is currently undergoing a paradigm shift, moving away from animal testing, and promoting mode-of-action based *in vitro* methods. In this context, genomics-based methods combined with an educated choice of biological systems have become attractive approaches due to their ability to generate readouts with high informational content, enabling educated hazard decisions. GARD™ (Genomic Allergen Rapid Detection) is a unique and highly versatile genomic-based *in vitro* testing platform using human cells that has the potential to accommodate many applications for assessment of varying toxicological endpoints. The term "platform" here indicates that all applications share the same experimental strategy and are based on the same modular building blocks comprising i) a biological system, ii) a training dataset of reference substances for data driven genomic biomarker identification, iii) a predictive genomic biomarker signature identified from genome-wide profiling, and iv) a prediction model, based on machine learning and pattern recognition algorithms. While the various modular building blocks of GARD™ have been thoroughly described for chemical sensitization (GARD™skin, GARD™potency, and GARD™air), it is important to appreciate that the individual modules are interchangeable and can be altered to target additional toxicological endpoints. This will be further demonstrated by showcasing current and future R&D projects targeting irritation and/or organ specific toxicity. To conclude, the performance demonstrated with the individual GARD applications indicates that the GARD platform is both a highly accurate testing strategy for assessment of chemical sensitization, as well as a highly promising tool for future applications."

[1]SenzaGen, Inc., USA; [2]SenzaGen AB, Sweden

Establishment of the Asian Consortium for Three Rs Supported by ASCCT

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In 2016, the Japanese Society for Alternatives to Animal Experiments (JSAAE) launched the first Asian Congress on Alternatives to Animal Experiments to promote throughout Asia the Three Rs—Replacement, Reduction, and Refinement—as guiding principles for a more ethical use of animals in scientific testing. Following the First Congress in Karatsu, Japan, during November 2016 and the Second Congress in Guangzhou, China, during October 2018, a Third Congress is now being planned for Korea during 2021. And to promote closer collaboration between our Asian colleagues, we are also now exploring the possibility of organizing for 2020 an Asian Consortium for Three Rs (Asian Consortium), which will provide funding and human resources in support of future Congresses in Asian countries.

JSSAE has concluded a memorandum of agreement with the European Society of Alternatives to Animal Testing (EUSAAT), the American Society of Cellular and Computational Toxicology (ASCCT), and the European Society of Toxicology in Vitro (ESTIV). Also, China's Toxicity Testing Alternative and Translational Toxicology (TATT) has concluded a similar memorandum with ESTIV.

Although our Asian colleagues are deeply involved in research and promotion of the Three Rs, countries in the EU and the USA are still the leading advocates for these principles, and we look forward to strong support for the Asian Consortium from our colleagues in ASCCT.

JaCVAM, NIHS, Japan and JSAAE

ASCCT 2019

Poster Abstracts

Prediction of a Vaccine Candidate for Neisseria Gonorrhoeae Adopting Reverse Vaccinology Approach

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Neisseria gonorrhoeae is causative of the sexually transmitted disease gonorrhea. As of now there is not yet a clinical approach that can prevent its infection. Hypothetically, the most promising approach would be to find one or more gonococcal vacines, but none has been successfully developed to the stage of validation. Therefore, as of now, management through antibiotic only is in practice. But, then, due to excess use of antibiotics, several resistant strains have arisen. Reverse vaccinology has been successfully applied to many infectious diseases. We applied a computational approach to identify and develop a new vaccine for reducing the burden of human suffering from gonorrhoeae. In the present study, reverse vaccinology approach has been adopted for the successful prediction of candidate vaccine targets in N. gonorrhoeae virulent strain NCCP11945. Proteome set was retrieved for this strain of N. gonorrhoeae, which contains 2163 proteins. Out of them, 37 trans-membrane proteins were screened using CELLO and PSORTb. Among these 37 proteins, 11 were identified by VaxiJen v 2.0 as the potential antigenic proteins. In the further screening nine proteins were identified as potential vaccine candidates. Among them one surface-exposed novel antigenic epitope "VQYAPKDNS" of membrane protein WP_012504064.1 was predicted to induce both B-cell and T-cell immune responses. This peptide can be used for the development of exclusive peptide vaccines. This being a humble beginning, the approach requires further designing of the vaccine and subsequent validation of the peptide target.

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Stakeholder Collaboration to Advance Human-Relevant Nonclinical Methods for Drug Development in the United States

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Recent initiatives in North America, such as the United States Food and Drug Administration's (FDA) Predictive Toxicology Roadmap, represent a shift in the way drugs will be developed and regulated. Regulators now state the need to integrate modern tools that better predict human outcomes than the traditionally used animal tests. In support of such initiatives and with the goal of increasing the human relevance of nonclinical drug testing, a growing group of professionals from federal agencies, the private sector and patient, health and research organizations collaborate under the Nonclinical Innovation and Patient Safety Initiative (NIPSI).

Through a *Drug Discovery Today* publication, NIPSI outlined factors that impede integration of new approaches and provide recommendations for addressing these factors. Ongoing projects focus on changing policy, supporting human-based science, and offering industry and regulator training. One policy project involves changing FDA and International Council on the Harmonisation of Technical Requirements for Pharmaceuticals for Human Use (ICH) regulations from requiring "animal" data to "nonclinical," which encompasses animal *in vivo* and human and animal-based *in vitro* and *in silico* approaches. Another project aims to establish an evaluation pathway for regulatory acceptance of human biology-based nonclinical approaches at FDA.

Physicians Committee for Responsible Medicine

California Cruelty-Free Cosmetics Act Establishes First North American Law Banning the Sale of Animal Tested Cosmetics and Ingredients

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The United States Food and Drug Administration does not require or review safety information prior to the marketing and sale of cosmetics in North America. Instead, individual companies are responsible for ensuring their cosmetic products are safe for human use. In 2018, the state of California joined the European Union and other regions in banning the sale of cosmetic products or ingredients that have been tested on animals, with limited exceptions. Starting January 1, 2020, no cosmetic may be sold within the state if it, or its ingredients, were tested on animals after January 1, 2020. Enumerated exemptions allow for testing conducted in response to a requirement by a state, federal or foreign regulatory body under certain conditions. However, the law also requires companies selling cosmetics or ingredients tested on animals under one of the exemptions must also use nonanimal methods to substantiate the safety of the product. Because the law applies to cosmetic companies as well as any third party suppliers or contractors, many ingredient manufacturers will be required to begin using *in vitro* or *in silico* methods, even if they are also conducting an *in vivo* study for the same endpoint to support the ingredient in another sector. This poster will outline the law and describe approaches available for use to meet California's legislative requirements.

Physicians Committee for Responsible Medicine

Abstract Sifter: A Literature Informatics Tool for Computational Toxicology

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The biomedical literature contains an abundance of information about the activity of chemicals in biological systems. The goal of literature informatics research at the EPA's National Center for Computational Toxicology is to use the literature more effectively in computational toxicology. To this end, we have developed a novel approach to article retrieval in our Abstract Sifter application. The Abstract Sifter is a document retrieval tool that integrates the richness of PubMed and other bibliographic sources with the powerful data-handling capabilities of Microsoft Excel. Results from searches are imported directly into an Excel sheet where the end-user can then use a novel "sifting" methodology for quick, agile relevance ranking of articles. The tool also enables article triage capabilities through easy tagging and noting functionality. Triaged citations can be exported to external software such as reference management tools. The Abstract Sifter can also provide a high-level view of a corpus of literature for a defined set of entities such as chemicals. This "landscape" view helps researchers assess the volume of literature in any given subject area to help with project scoping and chemical ranking and prioritization. Queries developed from the OECD Adverse Outcome Pathway (AOP) project connect key events in AOPs to the literature for chemicals on the Landscape sheet, offering evidence for inferring and investigating a chemical's mechanisms of action. The Excel format of the tool provides ease of use and facilitates collaboration. This abstract does not necessarily represent U.S. EPA policy.

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Facilitating Chemical Evaluation and Data Exploration with the Integrated Chemical Environment

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Currently, toxicity testing utilizes a complex suite of approaches to understand the interactions between test substance and target organism. Traditionally, such evaluations have relied upon results from *in vivo* models; new approaches can now integrate *in vitro* testing, *ex vivo* testing, *in silico* predictions and computational tools to inform the decision process. These approaches require readily available, well-characterized data that is relevant to the toxicity under investigation. Launched in 2017, the Integrated Chemical Environment (ICE) is the source for curated and toxicologically relevant data from the NTP Interagency Center for the Evaluation of Alternative Test Methods (NICEATM) and federal agency stakeholders and partners. To better support the needs of method developers and risk assessors, ICE updates have included new features aimed at making data exploration easier. In this presentation, we describe changes to the search function and other computational tools targeted to toxicologists and risk assessors who may not have ready access to computational toxicology resources. Key among the updates is the expanded *in vitro* to *in vivo* extrapolation (IVIVE) tool that allows users to access some of EPA's httk models using the ICE web interface. A simple case study will show how to retrieve Tox21 data relevant to *in vivo* endpoints and apply IVIVE workflows and other ICE tools. The case study will emphasize the applicability of ICE for a practical use case, demonstrate ease of use, and provide suggestions for data interpretation. ICE was supported with federal funds from the NIEHS, NIH under Contract No. HHSN273201500010C.

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Chemchart - Intelligent Platform for Chemical Data Management

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ToxTrack Inc is proud to unveil Chemchart – a flexible, intelligent platform for managing chemical data. Chemchart provides a single solution for managing the entire organizational chemical space. Embedded with machine-learning, Chemchart combines database management, document processing, and chemical exploration into an intuitive interface.

Chemchart combines big data flexibility and machine learning precision. Built for expansion, Enterprise's internal database easily extends to new types of data. Supporting the database is the most versatile search engine on the market – facilitating semantic and structural queries. Collections of chemicals can be easily compared.

Chemchart also has the ability to identify and extract chemical references in documents. The platform scans internal document collections (e.g. PDFs, spreadsheets, Word Docs, SDFs) and automatically indexes chemicals into the central database. In addition to organizing internal document repositories, Chemchart can join internal data with external datasets like patents, regulatory, or news repositories. Imagine getting alerts whenever a chemical in your supply chain lands on a new regulatory document, or finds a new use in a patent.

Go here to see Chemchart in action: https://www.youtube.com/watch?v=yZT3GnRpQZw

ToxTrack

Predictive Multitask Deep Learning Modeling of Estrogen Receptor Activities

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Endocrine disruption is a vital toxicity mechanism for many chemicals, especially those of environmental interest, such as pesticides. Traditional experimental testing, both *in vitro* and *in vivo*, to identify toxicants that can induce endocrine disruptions are expensive and time-consuming. Computational modeling is a promising alternative method for chemical toxicity evaluations. The rapid generation of data obtained from high-throughput screening (HTS) assays and computational power increase advanced computational modeling into a big data era. New modeling approaches, such as deep learning, are being used for model development. This work features an extensive modeling study using classic machine learning algorithms, normal deep neural networks, and multitask deep neural networks for 18 ToxCast estrogen receptor (ER) binding assays. Chemical descriptors consisted of two binary fingerprints (FCFP6 and MACCS keys). Each descriptor set was combined with four traditional machine learning algorithms (Bernoulli Naïve Bayes, k-Nearest Neighbors, Random Forest, and Support Vector Machines), two deep learning approaches (normal deep neural networks and multitask deep neural networks) to develop models for all 18 ER endpoints. The resulted model performance was evaluated using the area under the receiving operating curve (AUC). The results showed that individual models have AUC ranging from 0.57 to 0.86. External validation was conducted using new compounds collected from the Collaborative Estrogen Receptor Activity Prediction Project (CERAPP) with known *in vitro* ER agonist, antagonist, and binding activities as well as compounds from the Endocrine Disruptor Knowledgebase (EDKB) with known *in vivo* ER binding activity. An agonist, antagonist, or binding score was determined for each compound by averaging its predicted probabilities in relevant assays to compare to its known *in vitro* or *in vivo* activity. The results yielded AUC values ranging from 0.63 to 0.90 for *in vitro* outcomes and 0.81 to 0.88 for *in vivo* outcomes. We concluded that multitask deep neural network has advantages with large databases with mechanism-related toxicity endpoints (i.e., ER bindings), including in situations when using high-dimensionality chemical descriptors to extrapolate to *in vivo* outcomes. It is expected that this modeling strategy can be used for other computational toxicology modeling studies, especially for toxicity pathway modeling.

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Multi-Behavioral Endpoint Testing of an 87-Chemical Compound Library in Freshwater Planarians

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There is an increased recognition in the field of toxicology of the value of medium-to-high-throughput screening methods using *in vitro* and alternative animal models. The asexual freshwater planarian Dugesia japonica is an invertebrate model particularly well-suited for the study of developmental neurotoxicology. Our automated screening methodology allows for fast screening of multiple behavioral endpoints, developmental toxicity, and mortality. Using an 87-compound library provided by the National Toxicology Program, consisting of known and suspected neurotoxicants, including drugs, flame retardants, industrial chemicals, polycyclic aromatic hydrocarbons (PAHs), pesticides, and presumptive negative controls, we evaluated the benefits and limitations of the system for medium-throughput screening. We showed that, in the context of this library, planarians are the most sensitive to pesticides with 16/16 compounds causing toxicity and the least sensitive to PAHs, with only 5/17 causing toxicity. By testing diverse behaviors in both adult and regenerating/developing planarians, our screening platform uniquely allows differentiation of selective neurotoxic effects and effects specific to the developing nervous system which were observed for 13 chemicals in this library, including the known developmental neurotoxicant, chlorpyrifos. Furthermore, using Ward's method of clustering, we were able to differentiate some class-specific toxic profiles, including for the pesticides and flame retardants. Of the 87 chemicals, 28 had previously been evaluated in mammalian developmental neuro- (DNT), neuro-, or developmental toxicity studies. Of these 28, 18 had previously been identified as causing DNT in mammals and 20 were hits in regenerating planarians. This and similar studies will define the context of use for this new invertebrate system as a complement to other alternative models and computational tools to assay a larger swath of chemical and biological space with minimal time and cost.

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Alternative Vehicles Allow the LuSens Test to Predict Dermal Sensitization of Mixtures

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Predicting dermal sensitization is an important component of the acute toxicity testing battery. The second key event (KE2) in the dermal sensitization Adverse Outcome Pathway (AOP) is characterized by keratinocyte activation. The OECD 442D guideline details the *in vitro* keratinocyte ARE-Nrf2 Luciferase-based LuSens Test, herein referred to as the LuSens Test to predict UN GHS sensitizers (Category 1) or non-sensitizers. We performed a series of LuSens Tests using our validated protocol under the OECD 442D guideline. One limitation of this methodology for testing chemicals and mixtures is the limited solubility of potential test substances in the guideline-approved vehicles, Dimethyl Sulfoxide (DMSO), Media, and Water. To expand its applicability domain, we sought to validate the use of additional vehicles in the LuSens Test. We demonstrated eight additional vehicles to be compatible in the LuSens Test, as determined by their ability to appropriately promote positive and negative control luciferase induction responses. Using DMSO or Propylene Glycol the LuSens Test was able to predict the dermal sensitization potential of nine commercially available mixtures. Dermal sensitization predictions agreed in six of six mixtures tested where a safety data sheets were available. For two of two lotion mixtures tested, which were not expected to induce a sensitization response, no induction was seen. Lastly, a shampoo mixture had a positive prediction in both the LuSens Test and KE3 guideline-based assay. Together, these data demonstrate that the LuSens Test can be performed on mixtures using an expanded list of intra-laboratory validated alternative vehicles.

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Classification of EPA Ocular Irritants and Non-irritants by the OptiSafe™ Test Method

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The EPA eye irritation classification system is routinely used to categorize ocular toxicity. This system classifies chemicals that damage the eye after 24 hours (Category III, II, or I) and those that do not cause damage (Category IV). This latter classification is aligned with the standard definition of an "ocular non-irritant. The OptiSafe™ test is a novel, shelf-stable, *in chemico* method that can be used to discriminate ocular irritants/corrosives from non-irritants without use of animal tissues or cells. OptiSafe™ measures ocular damage via a proxy for the corneal stroma (water-soluble molecules), damage to phospholipid bilayers, and the potential to induce pH extremes in a system (pH buffering system of the eye). Chemicals in this study were selected based on a wide range of EPA classifications, chemical and physical properties, high quality *in vivo* reference data, and chemical stability. Selected chemicals (37) including surfactants not previously tested were aliquoted into coded vials and tested blind in triplicate. The coded vials were tested, and results were reported as either EPA Category IV (nonirritant) or not (EPA Category III, II, or I). The OptiSafe™ test method applied to these 37 test chemicals achieved a sensitivity of 100% (26/26), specificity was 81.8% (9/11), and overall accuracy was 94.6% (35/37). These results suggest that OptiSafe™ may be an important tool in the complete classification of eye hazards, especially for surfactants, as well as cosmetics and other substances applied to or around the eye.

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Use of Porcine Corneal Opacity Reversibility Assay (PorCORA) to Determine the GHS Classification for an Irritating Mixture (Cleaner Emulsifier and Degreaser)

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The PorCORA is a 21-day *ex vivo* ocular assay, which can distinguish between a substance's potential to cause severe (reversible) versus corrosive (irreversible) damage. We used the PorCORA's ability to determine GHS category 1 versus category 2 (2A/2B) of a Fryer Cleaner and Degreaser (FC-D) mixture formula. The bovine corneal opacity and permeability (BCOP) *In Vitro* Irritancy Score (IVIS) for a prototypical FC-D was 24.3 and its amine-based cogeners ranged between 35.5 and 50.8. However, the BCOP assay does not directly address reversibility of ocular damage, which is a key component of the GHS classification scheme for ocular irritants. The PorCORA revealed complete healing of the cornea by study day 7, indicating reversible eye irritation and non-corrosivity. The reversibility of damage for this material suggests that including the PorCORA can more accurately distinguish between GHS Category 1 and GHS Category 2 ocular irritation classifications than the BCOP alone. Thus, the PorCORA, along with other alternative test methods for ocular irritation such as the BCOP, can be used to assess Cleaner and Degreaser class mixture formula induced eye damage as well as the ability of this damage to heal, without the use of live animal testing.

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Increasing Confidence in the EASA Skin Sensitization Assay Using a Measurement Science Approach

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NICEATM is coordinating a multi-laboratory validation study to characterize the Electrophilic Allergen Screening Assay (EASA) for risk and hazard assessment for skin sensitization classification. The EASA assay functions by assessing potential skin sensitizers by measuring depletion of one of two probe molecules. The use of this assay in a single cuvette format resulted in multiple measurement challenges, such as low throughput and inability to include sufficient control measurements. We redesigned this assay protocol to work with a 96-well plate format. Process control measurements were incorporated into a 96-well plate design to quantify key sources of variability each time the assay is run. Additional control experiments were performed to evaluate photodegradation of probe molecules, bias from bubbles caused by pipetting, and statistical power of the assay system. Sixty compounds from NTP were tested in the 96-well plate design. One key insight revealed by this process was the interference from test compounds, namely producing an absorbance or fluorescence signal similar to that of the probe molecules, which would not have been previously detected using the single cuvette assay. Not taking the interference into account has been shown to lead to potential false negative identification. The data from the 60 compounds tested has approximately 75 – 80% concordance with LLNA data. Overall, the measurement science approach described here provides steps that can be taken to increase confidence in *in vitro* assays by fully characterizing sources of variability and potential biases in the assay that will facilitate interlaboratory testing and standardization.

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Documenting International Acute Systemic Toxicity Testing Requirements to Facilitate Increased Acceptance of Nonanimal Approaches

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The acute systemic toxicity tests – oral, dermal, and inhalation LD50 tests and variants – are the most common type of toxicity tests conducted worldwide despite ambiguous scientific relevance. In order to ensure that replacement test methods and strategies are accepted, it is important to understand the testing and information requirements of a variety of agencies covering a number of product sectors. While basic testing methods do not vary widely among countries, specific regional regulations, use contexts driving the conduct of these tests, and the degree of acceptance of alternative approaches are diverse. Understanding international requirements will inform scientific development and necessary regulatory policy changes to facilitate the reduction and replacement of animals in safety testing. Here, we present results from an international survey conducted with Organization for Economic Co-operation and Development (OECD) member and partner countries to identify specific test methods required and accepted for acute systemic single-dose studies within various product sectors (e.g., pesticides, industrial chemicals, pharmaceuticals, consumer products, and cosmetics). The survey also assessed acceptance of alternative approaches such as read-across, (Q)SAR, *in vitro* methods, and the potential for waiving tests or a combination of approaches. Results include not only frequently collected information by regulators but reported uses of information used for regulatory decision-making, which could serve as an information source to facilitate the development of alternative approaches. The presentation will provide recommendations to improve the implementation of alternative approaches within domestic regulations and identify areas for greater international harmonization.

Physicians Committee for Responsible Medicine

Phenotypic Profiling of the Acute Effects of Neuroactive Drugs in the Freshwater Planarian Dugesia Japonica

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Innovative computational approaches along with high-throughput screening (HTS) using *in vitro* and alternative animal models are revolutionizing drug discovery and toxicology. The asexual freshwater planarian Dugesia japonica is a promising invertebrate model with unique advantages for neurotoxicity HTS. Through quantitative assessments of morphology and multiple behaviors using a custom, robotic screening platform and semi-automated image analysis, we can differentiate the toxicity profiles of different chemicals. Here, we tested the acute effects of 9 neuroactive drugs, consisting of 3 anti-psychotics, 3 anti-depressants and 3 anxiolytics, on adult planarian nervous system function. Each drug was screened blind at minimum 5 different concentrations in half-log increments to evaluate dose-response. Additionally, negative and positive controls were tested. Each chemical was assigned a phenotypic profile based on the 15 endpoints tested, spanning body shape, unstimulated locomotion and stereotypical behaviors induced by light or temperature stimuli. Hyper- and hypo-active effects were distinguished to identify stimulatory versus depressive effects. Using hierarchical clustering, we were able to differentiate the three drug classes based on their phenotypic profiles alone. While small in scope, these results suggest that neuroactive compounds can be identified using this planarian HTS platform without knowledge of the molecular targets, which are generally difficult and time-consuming to identify. This systems level approach could thus be used to rapidly and cost-effectively screen small molecules by comparing the profiles of novel compounds to known neuroactive compounds. Using such a read-across based approach, we can better predict chemical effects to help streamline drug discovery and toxicology testing.

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Safety Assessment of the Use of Recycled High-density Polyethylene in Cosmetic Packaging Based on *In Silico* Modeling of Migration of EFSA Surrogate Contaminants and TTC for Dermal Sensitization and Systemic end-points

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A risk-based assessment of the safety of recycled high-density polyethylene (rHDPE) for use in cosmetic packaging was performed based on the guidelines published by the European Food Safety Authority (EFSA) to evaluate the use of recycled plastics for food. EFSA regulations require demonstration that the concentration of model chemical contaminants in recycled plastics is such that exposure from food use is less than the threshold of toxicological concern (TTC). We calculated the maximum concentration of EFSA model contaminants in cosmetic formulations (intended for both leave-on and rinse-off applications) which would not result in a daily exposure exceeding their respective TTC. Using mathematical modelling software (MIGRATEST© EXP), we then derived the concentration (Cmod) of each model contaminant in rHDPE which would result in the maximum concentration migrating from the rHDPE into each cosmetic formulation without exceeding the TTC. Cmod values were then compared to the EFSA reported concentration of each model contaminant (Cres) in the rHDPE. For each of the cosmetic formulations we evaluated the Cmod was less than the reported Cres. For skin sensitization we modeled a worst-case scenario and assumed 100% of each model contaminant in the rHDPE migrates into each cosmetic formulation. We then calculated the consumer exposure level for each contaminant based on dose per unit area and compared it to the dermal sensitization threshold (DST). The DST is based on the same principles as those used to develop the TTC to define a level of skin exposure where there is no appreciable risk of skin sensitization to an untested chemical. In each case we demonstrated that the migration of each model contaminant from rHDPE into each cosmetic formulation is far below the DST. In conclusion, this work demonstrates that rHDPE can be used safely for different cosmetic products.

Avon Products, Inc.

ChemAware Informatics Platform

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Introducing ChemAware: a new tool based upon DuPont's METIS chemical informatics platform. ChemAware is capable of providing rapid access to chemical information including toxicological hazards, persistence & bioaccumulation, occupational exposure limits, government & regulatory, and other relevant data. It is used to facilitate work done by Product Stewardship & Regulatory, Chemists, Toxicologists and other researchers. ChemAware is expected to be used throughout the DuPont supply chain wherever chemicals are involved.

Fast aggregation and visualization of chemical data enables researchers to leverage big-data to explore various hazard scenarios, develop chemical assessments and support critical business decisions. The ChemAware SQL Server database includes, but is not limited to, data from the US EPA, NLM, ECHA, Japanese NITE and common regulatory lists, as well as pre-calculated results from a variety of QSAR models in the absence of empirical data. Regular refreshes keep the database EverGreen providing the most relevant information. Users experience near immediate access to aggregations from relevant but disparate data sources, saving the researcher hours to potentially months of manual data collection.

Our next generation tool is multi-tenant, residing in the Microsoft Azure Cloud, making it securely deployable across organizations and ultimately available to the public. Each instance of ChemAware is separately administered with the tenant owner responsible for routine uploads. As DuPont currently intends to make ChemAware publicly available and EverGreen, a new open tenant will be established and maintained with regular uploads form public-only data sources. Other ChemAware tenants can be instantiated and managed by organizations establishing a trust relationship through the Azure Cloud. The organizationally managed tenants will be able to upload and leverage internal data in the privacy of their own network domain.

DuPont

The First FAIR Review Platform

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FAIR – findable, accessible, interoperable and reproducible – principles are 'good practice' for creating data sets. The integration of these policies into literature review comes at a critical time. Academic publication is expanding at an unprecedented pace and traditional research methods are badly in need of an upgrade to keep up. Public skepticism of previously trustworthy outlets (media, government, etc.) makes the transparency enabled by FAIR indispensable for building trust in academic review.

It is no longer possible for researchers to read all of the publications in their field. Even relatively specialized fields like "medical device stent" generate thousands of publications per year. Unfortunately, this means that most scientists are only exposed to a small fraction of the available research, and usually in a very biased way. When researchers use search engines to find relevant data they may look at only a few results until they find something fitting for their own research. Review platforms provide a streamlined process to perform thorough review of any topic.

When any kind of review is done, the process behind that review remains inscrutable. Which articles did the researcher consider? Why did they discard some articles and keep others? Online review applications make high resolution review referencing possible and FAIR data practices add transparency and accountability.

This presentation will focus on the novel aspects of the review application sysrev.com which launched in summer of 2019.

Insilica

Comparison of the Coverage of Three Publicly Available Profilers for Chemical Reactivity

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Covalent binding between an exogenous electrophile and an endogenous nucleophile acts as the molecular initiating events for a variety of adverse outcomes including skin sensitisation and genotoxicity. Several software tools have been developed that contain collections of structural alerts (profilers) that help identify electrophilic reactivity potential. Here, we compared three publicly available profilers to 1) understand their scope and coverage, and 2) assess their performance relative to a manually annotated skin sensitisation dataset based upon expert judgement. The three profilers comprised sets of SMARTS patterns associated with the five established mechanistic reaction domains (e.g. Michael acceptor). Based upon F1 score, the Enoch profiler performed the best for four of the five reaction domains, with the two other profilers performing better for the SN2 domain. There were 22 instances where all three profilers agreed with each other but disagreed with the manual assignment. These inconsistences could be explained by either the presence of an alternative mechanism, or some required biotransformation. The Tox21 library was then screened using the three schemes and the consensus outcome, in terms of mechanistic domain, was reported to identify chemicals likely to elicit electrophilic reactivity and, therefore, be more likely to elicit a non-target specific response. ToxPrint fingerprints were derived and activity enrichments computed to compare the structural features identified for the skin sensitisation dataset and Tox21 chemicals for each consensus reaction domain. *In chemico* reactivity data is currently being generated to evaluate the possible expansion/refinement of the profilers.

This abstract does not necessarily reflect EPA policy.

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Exploring Potential Refinements to Existing Threshold of Toxicological Concern Values for Environmentally-relevant Chemicals

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The US EPA are mandated by the Toxic Substances Control Act (TSCA) to perform risk-based prioritisation of chemicals and, for high-priority substances, integrate toxicity and exposure information to develop risk evaluations. One approach under consideration for chemicals with limited chemical-specific toxicity data is a Threshold of Toxicological Concern (TTC)-to-Exposure ratio. Within the TTC paradigm, a chemical is assigned to a specific class based upon an evaluation of chemical structure. Here, TTC values derived using (sub)chronic NO(A)EL data from EPA's Toxicity Values database (ToxValDB) were compared with published TTC values from Munro et al (1996). Toxtree was utilised to assign 4554 chemicals with structural information to their respective TTC category: Cramer structural class I, II, III, containing genotoxicity alert, or acetylcholinesterase inhibitor. 114 (2.5%) chemicals were deemed to be not appropriate for TTC. Similar TTC values were derived from ToxValDB compared to the TTC values from Munro: Cramer I (37.3 compared to 30 µg/kg-day), Cramer II (34.6 compared to 9 µg/kg-day), and Cramer III (3.9 compared to 1.5 µg/kg-day). For Cramer classes I and II, the 5th percentile values were not statistically different between the two datasets; whereas, the class III values were different. Chemical features of the two class III datasets were evaluated to account for differences in TTC values. The TTC values derived by Munro et al (1996) were substantiated by the expanded dataset in ToxValDB, reaffirming the utility of TTC as a promising tool in a risk-based prioritisation approach.

This abstract does not necessarily reflect EPA policy.

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Collaborative Modeling Project for Predicting Acute Oral Toxicity

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With an increasing number of chemicals to assess for acute systemic toxicity potential and a lack of available *in vitro* approaches, *in silico* models provide an alternative to predict acute oral toxicity and bridge data gaps. NICEATM and the ICCVAM Acute Toxicity Workgroup organized an international collaborative project to develop *in silico* models for predicting acute oral toxicity. In total, 35 groups participated, submitting 139 predictive models built using a dataset of 11,992 chemicals. Models were developed for five endpoints: LD50 value, EPA hazard categories, GHS hazard categories, very toxic (LD50 < 50mg/kg), and non-toxic (LD50 > 2000 mg/kg). Predictions within the applicability domains of the submitted models were evaluated using external validation sets, then combined into consensus predictions for each endpoint, forming the Collaborative Acute Toxicity Modeling Suite (CATMoS). The resulting consensus models leverage the strengths and overcome the limitations of individual modeling approaches. The consensus predictions performed at least as well as the *in vivo* acute oral toxicity assay in terms of accuracy and reproducibility. CATMoS consensus models are available as free and open-source tools via the OPERA predictive tool (https://github.com/NIEHS/OPERA), which provides applicability domain assessments and accuracy estimates. CATMoS predictions for the ~850k chemical structures in DSSTox will ultimately be publicly accessible via NTP's Integrated Chemical Environment (ice.ntp.niehs.nih.gov) and the EPA's CompTox Chemicals Dashboard (comptox.epa.gov/dashboard). This project was funded with federal funds from NIEHS under Contract No. HHSN273201500010C. This abstract does not necessarily reflect EPA policy.

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Evaluation of the Predictive Accuracy of QSAR Models and Alerts for Genotoxicity Using a Newly Compiled Experimental Data Set

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Ratiometric β-lactamase (BLA) reporter assays have seen increased used high throughput screening (HTS) programs for chemical assessment. While these reporters are highly sensitive, customizable, and control for plate and well variability, the direct inference from the ratiometric BLA endpoint may be difficult to interpret. Consequently, chemical assessment and structure-activity models based on the data may be confounded by non-linear effects on different channels in the ratiometric set up. We fit and analyzed the concentration-response curves produced by the 10,000 chemicals screened in seven BLA stress-response assays as a part of the Tox21 initiative. Particular attention was given to the relationship between the three BLA assay readouts: background, target, and ratio; and uncertainty quantification. The ratio is often used solely for activity classification. However, we found that activity classifications based on a BLA ratio readout are confounded by interference patterns for as many as 85% of active chemicals in some assays. Furthermore, the potency and efficacy estimates derived from the ratio readouts may not represent the target channel effects and thus complicate chemical comparison. Therefore, we recommend a direct analysis of the target gene-expression channel to determine the target activity in a BLA assay followed by analysis of background fluorescence or viability counter screens to screen for loading or viability artifacts. The difference between viability and target gene effects is best assessed explicitly for each exposure experiment based on robust parameter uncertainties. This approach eliminates the channel interference issues and allows for straightforward chemical assessment and comparisons.

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CRapid Evidence Mapping for Health: A Case Study

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Environmental health stakeholders increasingly rely on systematic review tools and practices to summarize the literature and identify consensus regarding potential health risks. Given the ever-accelerating pace of publications in this field, the practice of "evidence mapping" is used to identify key areas of study relevant to a given topic along with gaps in the literature. However, constructing detailed evidence maps is resource-intensive, limiting their utility for practical implementation. Here we outline "rapid Evidence Mapping" (rEM), a resource-efficient form of knowledge synthesis in which components of the systematic review process are simplified to produce a visual and quantitative representation of the scientific evidence. We show how rapid evidence maps can be created with the aid of Sciome's text-mining and machine learning software, and describe a case study on the topic of low-calorie sweeteners (LCS) with respect to human dietary exposures and health outcomes. The resulting rEM produced findings similar to a traditional evidence mapping of the same topic (Wang et al. 2016) but required significantly less time and resources to create. Furthermore, a sensitivity analysis evaluating the set of studies included at 25% recall (i.e., the point at which the machine learning algorithms predicted we had identified 25% of all relevant references) would have resulted in the same conclusions, suggesting that further efficiency gains can be achieved by mapping only a subset of the available literature. The potential time savings of the rEM approach make it a powerful tool for rapidly translating knowledge to inform science-based decision-making.

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Suitability of BioOcular® *In Vitro* Tissue Model for Eye Irritation Potential in Color Cosmetic and Sunscreen Formulations with Varying Product Forms and Compositions

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OBJECTIVES: BioOcular® is a reconstructed human cornea-like epithelial 3D tissue model developed in China. Previously, we demonstrated promising acceptability of the BioOcular model to predict eye irritation potential for facial and eye area products. Here, we aim to evaluate the suitability of the BioOcular® tissue model to assess eye irritation in other cosmetic products such as color cosmetic and sunscreen formulations with varying product forms and compositions. MATERIALS AND METHODS: Twenty-one cosmetic formulations, including five solid facial powders, five liquid facial makeups, six solid eyeshadows, one liquid eye shadow, two lipsticks and two sunscreen lotions were tested. The BioOcular® cultures were treated in duplicate with neat test articles at four exposure times up to 24 hrs. 0.3% Triton™ X-100 was used as positive control and sterile, deionized water was used as negative control. RESULTS: Most tested color cosmetic formulations resulted in an ET50 of > 24 hrs except for one liquid facial makeup with an ET50 of 6 hrs; The sunscreen lotions had an ET50 of > 24 hrs and 6.8 hrs, respectively. CONCLUSIONS: The BioOcular® tissue model has shown to perform, and be suitable to, assess eye irritation potential of different forms and compositions of color cosmetic and sunscreen formulations. Most of the color formulations, except for one liquid facial makeup, were tested for the first time at Zhejiang Institute for Food and Drug Control. Based on the obtained ET50s, the tested color cosmetic formulations are ultra-mild in nature, as expected. This study further demonstrates the usefulness of the BioOcular assay for test articles in the ultra-mild range, and as an emerging model in China.

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Co-expression Network Analysis to Identify Heterogeneity Between the Breast Cancer Cell Line MCF-7 and Human Breast Cancer Tissues

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Lack of characterization of cell lines can have serious consequences on the translatability of *in vitro* scientific studies to human clinical trials. This project focuses on the Michigan Cancer Foundation-7 (MCF-7) cells, a human breast adenocarcinoma cell line that is commonly used for *in vitro* cancer research, with over 33,000 publications in PubMed. Previously, our center has shown that even MCF-7 cells obtained from the same cell batch at the same cell bank can display cellular and phenotypic heterogeneity, which affected reproducibility of experiments using this cell line. In this study, we explore the key similarities and differences in gene expression networks of MCF-7 cell lines and human breast cancer tissues. The MCF-7 dataset includes 1032 samples obtained from ARCHS4, and the human breast cancer tissue dataset came from the Cancer Genome Atlas, including 1098 breast tissue samples from individuals with breast invasive carcinoma. We used Weighted Gene Correlation Network Analysis (WGCNA) – a method that takes advantages of correlation amongst genes and graph theory – to explore similarities and differences in key transcription factors, signaling mechanisms and pathways in MCF-7 cell line and human breast cancer tissues. We found that although the majority of genes in the two networks were non-overlapping, there are some conserved subnetworks. Since cancer cell lines are commonly used in both basic and translational research to understand biological mechanisms, drug effects, and toxicology pathways, our comparison of the regulatory networks of MCF-7 and breast cancer tissues can address some concerns over the validity of using cancer cell lines as standard models for humans.

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Identifying Eye Irritants (GHS Category 2) Using Validated, Non-Animal Tests

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OECD Test Guideline 405 prescribes a weight-of-evidence (WoE) analysis and sequential testing strategy for the classification of acute eye hazards, including, "Perform validated and accepted *in vitro* or *ex vivo* ocular test(s)." Four tests qualify: three designed to identify severe eye irritation/corrosion (GHS Category 1) and one to identify non-irritants (GHS No Category). GHS Category 2 (eye irritant) classification is impossible using any single test. The EpiOcular™ Eye Irritation Test; EIT (OECD 492) classifies a substance to be No Category, or contrarily causes (uncategorizable) eye effects. Conversely, the Bovine Corneal Opacity and Permeability (BCOP) Test (OECD 437) is used for ruling in or ruling out Category 1 effects. By using a dual-assay/approach system – the combination of the EIT and BCOP test – we have determined, with a high degree of accuracy, GHS Acute Eye Hazard Category 2 chemicals that cause reversible eye irritation. When a BCOP test rules out GHS Category 1, and the EIT rules out GHS No Category, analysis of these results indicates the only other possible designation – Category 2. Per GHS, Category 2 classification defaults to Category 2A, because differentiation between Category 2A and 2B cannot be made. After testing 42 chemicals, we correctly identified 93% of the Category 2A/B chemicals as Category 2. The potential of the BCOP EIT dual-assay system, coupled with WoE evaluation, to correctly classify substances into GHS Category 1, Category 2A, and No Category is encouraging. We predict using this testing strategy would greatly reduce reliance on Draize Rabbit Eye Tests.

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Ethanol as an Alternative Vehicle for Determining Skin Sensitization Potential Using the Human Cell Line Activation Test (h-CLAT) Assay

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Proper identification and classification of skin sensitization potential for new consumer products, chemicals, and pharmaceuticals are important for hazard communication and risk management, upon which *in vitro* toxicology methodologies heavily rely. h-CLAT, uses dendritic cell proxy THP-1 monocytic cell line to address the third key event (dendritic cell activation) in the sensitization Adverse Outcome Pathway, during which time the cell surface markers CD86 and CD54 are induced. If a test article (a volatile liquid fragrance) induces CD54 and/or CD86 expression to a level of at least 200 and 150 RFI, respectively, it is predicted to be a skin sensitizer. A major limitation to the versatility of h-CLAT is the few recommended vehicles: saline, media, or dimethylsulfoxide. However, OECD 442E allows alternative vehicles with sufficient scientific rationale. Here, the test article was not soluble in the recommended vehicles; therefore, we sought to determine a suitable alternative and provide sufficient scientific rationale for its use. The test article was soluble in ethanol up to the recommended 100 mg/ml concentration, and the h-CLAT was performed. Positive (2-mercaptobenthiazole) and negative (isopropanol) controls were concurrently tested using ethanol as their vehicle. h-CLAT correctly predicted the negative control not to be a skin sensitizer, and the positive control as a skin sensitizer. The test article was predicted to be a skin sensitizer, with the RFI levels of CD54 and CD86 induced above 200 and 150, respectively. Here we provide scientific evidence and rationale for ethanol to be used as a suitable vehicle with the h-CLAT method.

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Use of an Alternative Vehicle in the Human Cell Line Activation Test (h-CLAT) to Broaden the Utility of the Test to Detect Dermal Sensitizers

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The h-CLAT was developed as an *in vitro* method of identifying skin sensitizers and was promulgated by OECD as a testing guideline in the spirit of advancing the 3Rs (Replacement, Reduction and Refinement). The guideline test is limited by the sanctioning of only two currently validated vehicles (DMSO and saline), precluding *in vitro* sensitization testing of a wide range of insoluble chemicals. One such insoluble test chemical, Tetrakis (2-ethyl butyl) Orthosilicate, was evaluated in the h-CLAT using an alternative vehicle (i.e., ethanol) in an effort to broaden the utility of the test. This test measures the selective induction of the surface markers CD54 and CD86 in the human monocytic leukemia cell line THP-1, which functions as a dendritic (Langerhans) cell surrogate. Known reference chemicals such as mercaptobenzothiazole (MBT), used as a positive control, and isopropanol, used as a negative control, were also included to evaluate the utility of ethanol as an alternative h-CLAT vehicle. MBT induced positive responses for CD54 and CD86 expression as expected, whereas isopropanol resulted in negative responses for both cell-surface markers. The test chemical was found to be negative in this test.

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Use of Porcine Corneal Opacity Reversibility Assay (PorCORA) to Evaluate the Hazard of a Concentrated Laundry Detergent

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Through collaboration between SC Johnson and MB Research Labs (both of whom have a long history of using alternative methods to reduce dependency on animals in toxicity testing) a concentrated laundry detergent product mixture was evaluated in the porcine corneal opacity reversibility assay. The PorCORA is an *ex vivo* assay developed to predict the reversibility (healing potential) of ocular irritants. Historical bovine corneal opacity and permeability (BCOP) data classified the formula as a GHS Category 1 eye irritant. However, the BCOP assay does not address reversibility of ocular damage, which is a key component of the GHS classification scheme for ocular irritants. PorCORA results indicated that the ocular damage induced by exposure to the concentrated laundry detergent mixture was fully reversible. The inclusion of reversibility of damage as an endpoint suggests that the PorCORA can supplement BCOP assay results to distinguish between GHS Category 1 and GHS Category 2 ocular irritation classifications. Thus, the PorCORA, along with other alternative test methods for ocular irritation, can be used to assess consumer detergent product-induced eye damage as well as the ability of this damage to reverse (heal) following detergent mixture exposure to the eye without the use of animal testing.

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Comparison of *In Vitro* 3D Reconstructed Human Epidermis Models for the Skin Irritation Test with Surfactant-Based Formulations

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Objective: Understanding skin irritation potential is essential in evaluating the safety of cosmetic formulations. Our study compares three *in vitro* 3D reconstructed human epidermis (RhE) models in evaluating skin irritation of surfactant-based rinse off products. Methods: Three formulations with surfactant concentrations of 7.56%, 12.56%, and 17.56% were tested utilizing EpiDermTM provided by MatTek, EpiKutis® provided by Guangdong Biocell Biotech Co. and SkinEthicTM RHE provided by Shanghai EpiSkin, following the methodology described in OECD Test Guideline 439 for evaluating skin irritation potential. A test article with 7.56% surfactant and spiked with 10% Triton X-100 was also included as a matrix positive control. To note, EpiKutis® and SkinEthicTM are RhE models originating from Chinese skin. Results: All three models differentiated surfactant concentrations in a dose-response manner as expected. The higher the surfactant concentration, the lower the ET50 value observed. In general, EpiKutis® and SkinEthicTM models consistently showed lower ET50 values than the EpiDermTM model across formulations. This difference was not significant as to reclassify results. Conclusion: All three models demonstrated equivalency in predicting skin irritation potential. ET50 values were comparable and within expected results range. Minor variabilities observed could be explained by difference in tissue structure or system standard testing conditions. While further evaluation of more product categories is needed, our study supports the opportunity for *in vitro* models to be used more broadly within the cosmetic industry in China.

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Promoting the Uptake of Alternatives to Animal Testing Through the Development of eLearning Tools

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In order to further promote the implementation of Directive 2010/63/EU, the European Commission issued calls for a number of related projects last year. One of these projects is aimed at facilitating the uptake of non-animal alternatives by developing two e-learning modules. The contract for this project was awarded to a consortium consisting of SYRCLE, the Swiss 3R Competence Centre, lnstitute for ln Vitro Sciences, Pharma Launcher and Ecorys UK. This consortium will develop two modules, i.e., one e-learning module focussed on searching for existing non-animal alternatives (including systematic reviews) and one module targeted at researchers who want to develop reliable and relevant non-animal alternatives for regulatory use taking into account Good In Vitro Method Practices (GIVIMP). The quality of the developed modules will be assessed by external review groups. The learning outcomes will be presented to the commission along with the design of the assignments through which these outcomes will be realized.

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ASCCT Awards

One of the main aims of the ASCCT is to support and engage young scientists working in the *in vitro* and computational toxicology fields. To do this we provide financial awards and travel bursaries to our annual meeting as well as other topical meetings.

Edward Carney Predictive Toxicology Award

In 2015, in memoriam of Dr. Edward Carney, the Society established the Edward Carney Predictive Toxicity Award. Dr. Carney was an active and dedicated member of the ASCCT, and a partner, mentor, and friend who inspired many in our fields. The award is $500, and will be awarded to a winning first author presenting at each annual ASCCT meeting, to assist with travel and/or research expenses.

Previous Award Winners

2015: Dr. Nicole Kleinstreuer: Identifying reference chemicals for androgen receptor activity

2016: Emma Bowers: Modeling a com *yap.4@wright.edu* plex in vivo response in vitro: Exploring heterogeneity and mechanisms associated with ozone adaptation

2017: Ellen Garcia: Characterization of two lung cell lines for use in cell division focused, single-cell toxicity assays

2018: Dr. Sudin Bhattacharya: Integrating Genomics and Epigenomics Into Predictive Toxicology of the Aryl Hydrocarbon Receptor

Tox 21 Student Award

After winning the William and Eleanor Cave Award for his career achievements in in vitro toxicology, Dr. Ray Tice generously established the Tox21 Student Award. Under this award, $500 will be given at each annual meeting for the next five years for the best student presentation.

Previous Award Winners

2016: Ellen Garcia: Single-cell analysis reveals that silver nanoparticle exposure leads to multi-nucleation through defective cell division

2017: Wenyi Wang: Mechanistic evaluation of chemicals that induce oral acute toxicity by mitochondrial membrane disruption: Big data profiling and analysis

2018: Daniel Russo: Developing Mechanism-Based Animal Toxicity Models: A Chemocentric Approach Using Big Data

2019 ASCCT Notes

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