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COMPILATION OF ABSTRACTS

Platform Presentation Abstracts

Session 1: Toxicology

WHOLE ANIMAL TOXICOLOGY SCREENING ON THE FLY

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A major problem facing the field of toxicology is how to test the tens of thousands of synthetic chemicals integrated into the fabric of human life from food additives to building materials. Here, we present an in vivo model using *Drosophila melanogaster* to test the effect of chemicals on stem cells and the stem cell microenvironment. As a proof-of-principle, we screened 80 compounds from the National Institute of Environmental Health Sciences' Tox21 project. We fed these compounds to flies over a period of three days, dissected their intestines, and used confocal microscopy to detect the effect of each on the growth and health of the GFP-marked intestinal stem cells. Of the 80 compounds screed, 10 caused death of adult fruit flies. This was a reassuring finding, as all 10 are pesticides currently in use either worldwide or specifically in the United States. When these insecticides were diluted to sub-lethal levels, they had no effect on stem cells. However, of the remaining 70 non-insecticides, we found one had a stimulatory effect on stem cells, resulting in small tumor-like growths. This is of great concern because the compound, methylmercury, is often found in our food chain, especially in fish meat. Prior to our findings methylmercury was known predominantly for its effects as a neurotoxin. To explore whether the growth promoting effects may also be expected in mammals, we tested whether it acted through the evolutionarily conserved JAK/STAT inflammation and JNK stress response pathways. Using a reporter for JAK/STAT signaling, we found that methyl mercury induces cells in the stem cell microenvironment to secret the JAK-STAT ligand unpaired 3. Furthermore, using tissue-specific RNAi knockdown of the JAK/STAT dome receptor, we found that stem cells must receive unpaired-3 ligand to respond to methylmercury insult. These results demonstrate the utility of *Drosophila* as an in vivo screening tool, and highlight a previously unrecognized mechanism by which methylmercury may be toxic to animals. Additionally, our results suggest that methylmercury may act as a carcinogen.

INVESTIGATING THE ECOLOGICAL AND EVOLUTIONARY EFFECTS OF CHEMICAL STRESSORS USING MUMMICHOGS

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The USEPA uses adverse outcome pathways (AOPs) as a conceptual framework to drive the utilization of molecular and physiological pathway-based data in assessing the hazards of stressors to human health and wildlife populations. Mummichogs (*Fundulus heteroclitus*) are useful study organisms to support AOP development due to the availability of well-developed molecular resources and their effectiveness in linking controlled laboratory experiments to field observations for development of ecological models. To understand the effects of chemicals and other stressors on population persistence, we are developing and testing models of fecundity and dynamic energy budgets, which will ultimately be integrated into individual-based models of population persistence. To support the development of these models we used small groups of individually tagged mummichogs to investigate the effects of dietary exposure to a variety of stressors (i.e., mercury, flame retardant bis(2-ethylhexyl)-2,3,4,5-tetrabromophthalate (TBPH), and polychlorinated biphenyls (PCBs), and resource limitation) on mummichog physiological condition, growth, and

reproduction. Furthermore, we took advantage of locally adapted populations of the non-migratory mummichog to investigate the evolutionary consequences of long-term pollutant exposure. Using populations of mummichog that have evolved tolerance to severe industrial pollution, including to (PCBs) and polycyclic aromatic hydrocarbons, we used quantitative trait loci (QTL) studies and population genomics to identify the specific genetic changes underlying adaptation to these chemicals. Using laboratory-based growth and breeding studies, we are also investigating the potential energetic consequences of this evolved tolerance. Overall, the combination of laboratory experiments, field study, and ecological models provides a testable system to investigate and predict the effects of stressors on individuals and populations, and to connect those effects to underlying molecular mechanisms.

SITE-SPECIFIC RISK ASSESSMENT OF PETROLEUM PHOTOTOXICITY USING THE PHOTOTOXIC TARGET LIPID MODEL

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Solar radiation increases oil toxicity considerably through phototoxic action. This occurs when certain phototoxic components in the oil mixture, including parent and alkylated polycyclic aromatic hydrocarbons (PAHs) and benzothiophenes, absorb light in the ultra violet (UV) and visible (VIS) portions of the solar radiation spectrum after being bioconcentrated in the target lipid of marine organisms. This paper proposes a method to predict site-specific phototoxic risk for oil. Accordingly, a phototoxic target lipid model (PTLM) of PAHs' acute phototoxicity will be presented that is based on the target lipid model (TLM) of PAHs and other narcotic chemicals. It can predict either the median lethal concentration (LC50) measured at a fixed duration of irradiance exposure or the LT50, the time required to achieve 50% mortality at a fixed concentration. The model accounts for all factors involved in phototoxicity, including physiochemical properties of the chemicals (octanol-water partitioning and the molar adsorption spectra of the PAHs of concern) and the ambient conditions (the spectra of the light irradiance and the duration of exposure to light). The only species dependent parameter in the model is the critical target lipid body burden (CTLBB), which is obtained from the TLM. The organism lipid normalized body burden produces 50% mortality. Lipid normalized body burdens apply to any narcotic chemical and are currently available for 47 aquatic animals and five algal species. The PTLM is validated by application to 12 alkylated PAHs and other phototoxic chemicals in oil. It is applied to mixtures of PAHs by assuming additivity of the toxic units computed for each of the PAHs in the mixture. The results of the calibration and its application to mixtures of PAHs and to water accommodated fractions from various neat and weathered petroleum samples will be presented. Since the PTLM accounts for all factors involved in phototoxicity, its outputs can be used for site-specific petroleum phototoxicity risk assessment to replace the previously proposed generic phototoxicity thresholds.

DEVELOPMENTAL EXPOSURE TO XENOESTROGENS AND THEIR EFFECTS ON THE FEMALE MOUSE MAMMARY GLAND

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The mammary gland undergoes different phases of development in the embryonic, pre-pubertal, pregnancy, lactation, and involution stages of a mammal's life. During these time periods, the mammary tissue is strongly influenced by hormones; for this reason, scientists have hypothesized that synthetic chemicals with hormonal activities could disrupt

gland development and contribute to diseases like breast cancer. Bisphenol S (BPS) is an organic compound that is used in many everyday consumer products and human exposure is widespread. BPS has been used to replace Bisphenol A (BPA), which is known to affect the mammary gland. Ethinyl Estradiol (EE2) is a pharmaceutical used in contraceptive pills. Our study examines the effects of early life exposure to BPS or EE2 on the mammary gland of female mice at pre-puberty and puberty. To assess effects on the mammary gland, growth parameters in whole mount glands are quantified using volumetric morphometrics. The histological appearance of mammary glands is also assessed using standard histological methods; proliferation and protein expression is quantified using immunohistochemistry. These assessments will allow us to better understand whether environmentally relevant doses of two synthetic estrogens disrupt development of the female mammary gland. This study is especially important because although BPS exposures are widespread, this compound is poorly studied.

Session 2: Emerging Contaminants

PFAS DATA CERTAINTY ISSUES AND RECOMMENDATIONS TO OBTAIN ACCURATE DATA FOR RISK EVALUATIONS.

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Currently, EPA Method 537 is the only published regulatory method for analysis of per- and polyfluoroalkyl substances (PFAS) and this method is only applicable to drinking water. Therefore, the environmental community and regulatory decision makers face broad technical gaps and challenges to interpretation of PFAS contamination in environmental media such as groundwater, soil, sediment, and biota.

The Interstate Technology & Regulatory Council (ITRC) has gathered a PFAS team of scientific experts to develop guidance and produce a series of six Fact Sheets on the following core PFAS topics:

- History and Use of Environmental Sources
- Nomenclature Overview and Physicochemical Properties
- Fate and Transport
- Site Characterization Tools, Sampling Techniques, and Laboratory Analytical Methods
- Remediation Technologies and Methods
- Regulatory Summary

This presentation will focus on Fact Sheet #4 based on the author's direct participation on the ITRC PFAS team and additional analytical experience. Specific issues, limitations, and interferences in PFAS data using EPA Method 537 will be discussed along with the key factors affecting analytical accuracy of PFAS compounds in media other than drinking water. Differences in quantitation approaches (e.g., surrogates *vs*. isotope dilution techniques) and their effect on data certainty will be presented as well as recommendations to obtain technically valid data in environmental media important for evaluating exposure pathways.

ADVANCES IN POLY- AND PERFLUORALKYL SUBSTANCES (PFAS) ANALYTICAL TECHNIQUES: IMPLICATIONS FOR CONCEPTUAL SITE MODELS.

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Poly- and perfluoroalkyl substances (PFAS), including perfluorooctane sulfonate and perfluorooctanoate are commonly elevated in soil and groundwater at sites with industrial PFAS applications or past use of firefighting foams, including aqueous film-forming, fluoroprotein and film foaming fluoroprotein foams (AFFF, FP, and FFFP). The products contain a complex mix of fluorinated compounds that include perfluorinated compounds, where all carbons are saturated with F atoms, and polyfluorinated compounds where some carbons have hydrogen bonds. The polyfluorinated compounds, termed precursors, are transformed in the environment to form perfluorinated compounds which are

extremely persistent and not susceptible to further transformation. The precursors are not accounted for by the U.S. EPA analytical method 537; however, precursors represent a "hidden" mass that should be considered in fate and transport assessments and conceptual site models, especially given that many PFAS are anionic and are not retarded significantly in the subsurface, whereas some precursors are cationic and bind to soils via ion exchange mechanisms and represent a less mobile source mass. This presentation will discuss three new analytical methods developed to quantify the total concentration of precursors and PFAS in water and soil samples. The analytical methods include the total oxidizable precursor (TOP) method, particle induced gamma emission (PIGE) spectroscopy, and adsorbed organic fluorine (AOF). This next generation of PFAS analytical techniques are generating more comprehensive analytical data that supports more robust conceptual site models and are improving our understanding of PFAS fate and transport. Accounting for precursors is also key for the successful design of remedial systems.

PFAS IN MAINE WATER, SEDIMENT, AND FISH

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PFAS (poly- and perfluoroalkyl substances) were investigated within surface water, sediment, and fish tissue from two stream drainages originating at a former Air Force base. PFOS (perfluorooctanesulfonic acid), PFHxS (perfluorohexane sulfonate), and PFOA (perfluorooctanoic acid) were the most commonly detected PFAS in surface water. PFOS, PFHxS, and PFOA concentrations in surface water ranged from 0.47 to 392 ng/L, 1.55 to 371 ng/L, and 1.9 to 33.2 ng/L, respectively. PFOS concentrations exceeded the Maine Center for Disease Control (MeCDC) surface water screening levels at 2 of 17 stream and lake locations draining the base, but PFOA did not exceed any screening level. PFOS concentrations in 17 sediment samples ranged from 0.63 to 10.7 µg/kg (PFHxS and PFOA were below the reporting limit) and did not exceed MeCDC screening levels. PFOS was the most commonly detected PFAS in fish tissue samples (PFHxS and PFOA were mostly below the reporting limit) and concentrations ranged from 2.17 to 457 µg/kg. All fish tissue samples exceeded the subsistence fisher screening levels except the background samples. Most fish tissue samples (85%) exceeded the recreational fisher screening levels. PFAS measured in fish from some other Maine lakes and rivers ranged from non-detect to levels of potential concern.

Session 3: Contaminated Soils/Sediments

ASSESMENT OF THE BIOGEOCHEMICAL DYNAMICS OF MERCURY IN RIVER BANK SOILS (SOUTH RIVER, VA)

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South River (Waynesboro, Virginia) is a legacy mercury (Hg)-impacted site from an industrial source; the sediments beneath and adjacent to the river and soils in the floodplain contain elevated concentrations of mercury. The understanding of redox-driven transformations and speciation of mercury in soil and freshwater plays a crucial role in the ecosystem biogeochemistry and environmental fate of this toxicant. Sediment erosion—including the resuspension of Hg-bearing particles—is one possible source of mercury release into the river water. Changes in water level in the river (e.g. by precipitation) can cause changes in the flow regime in the adjacent soil and ground water zone, possibly influencing moisture contents and flow direction and velocity. These changes can impact the aqueous and solid-phase chemistry and may cause changes in mercury speciation and transport. In this field study, we examined the complex interactions between the riverbank and channel system, investigated the processes that govern biogeochemical transformation of mercury in the Hg-impacted soils, and characterized changes in geochemical gradient in soils over time. Sampling of the banks and adjacent flood plains as well as river channel was performed several years using conventional and novel sampling techniques. Passive sampling techniques, diffuse-gradient in gel-thin film (DGT) devices, were applied to measure concentrations of labile/dissolved mercury and methylmercury in porewater of bank soils and river sediment. Redox condition in the bank and channel were monitored by voltammetry to measure dissolved redox sensitive species, such as oxygen, iron, manganese, and sulfide. Results showed that mercury levels during high flow events exceed 10 times the baseline concentration. Multiple high flow events occur each year (with a repetition frequency of about 2 weeks). Bank leaching and groundwater flow may be a larger source of mercury to the river than currently accounted for in the conceptual model. Geochemical data showed flood events cause changes in redox conditions in the bank, possibly caused by bank flushing. These redox changes may enhance mercury release from the banks. Field collected chemical and groundwater physical monitoring data are used to make long-term projections of mercury and methylmercury flux and release as well as to improve conceptual models of mercury behavior in the South River.

1, 4- DIOXANE – A REVIEW AND EVALUATION OF THE AVAILABLE ANALYTICAL METHODOLOGIES USED IN SUPPORT OF THE LATEST STATE AND FEDERAL STANDARDS

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A widely used chemical compound, 1,4-dioxane has a multitude of industrial uses and may be present in a variety of commercial products. Historically, 1,4-dioxane has not been included on most laboratory or regulatory target compound lists. When it was included on VOC target lists, many of the physical and chemical properties that make it such a useful industrial chemical also made it very difficult to determine using conventional analytical methods.
Due to the relatively recent classification of 1,4-dioxane as “likely to be carcinogenic to humans” , many regulatory standards and guidelines have been lowered. This presentation will review the three most commonly used analytical methods for low level 1,4-dioxane analysis – Method 8260 SIM, Method 8270 SIM and Method 522. Each method will be evaluated and the data generated in a method comparison study will be presented.

IMPROVEMENTS IN THE SEDIMENT QUALITY TRIAD FOR METALS-IMPACTED SEDIMENT

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Designing a Sediment Quality Triad always looks good on paper but, more often than not, yield nebulous results. Confounding variables include variation in TOC and grain size (affecting both bioavailability and organism response), variable habitat with different benthic communities, and the presence of non-site/nonpoint pollutants. These studies are costly and often times a dose-response curve is not observed, especially when the contaminant concentrations are not distributed across a wide enough range of concentrations. At a Massachusetts site historically impacted by lead chromate, our conceptual model indicated that both lead and chromium would be sparingly soluble, so low bioavailability and low toxicity were anticipated (even well above the PEC). We developed a robust field and laboratory design to attempt to minimize methodological and regulatory uncertainty. Lead concentrations in the sediment ranged from low ppm in the reference brook to low percent levels in the impacted brook. Recognizing that randomly collecting sediment samples from impacted reaches would not necessarily provide a proper range of "doses" we 1) scrutinized historical data to determine locations that could provide a *wide* range of contaminant concentrations 2) collected samples at more locations than we intended to test in the bioassay 3) used additional 'chelation' replicates as an internal TIE control (to identify site-specific effects) 4) instructed the toxicity laboratory to store all the samples until the analytical results provided us with an optimal range of "doses" and 5) collected benthic macroinvertebrates immediately adjacent to samples obtained for sediments for toxicity testing. Although these improvements required more field effort, the improvements only added 15% to the total cost of the project and, more importantly, allowed us to choose the proper sediment levels to test, ultimately yielding a *robust range of dose-response concentrations.* The results validated the conceptual model, resulting in high confidence in the risk management strategy and greatly narrowing down the range of remedial options.

Session 4: Ecological Risk/Ecological Services

POPULATION-RELEVANT ENDPOINTS IN ECOTOXICOLOGICAL HAZARD AND RISK ASSESSMENT OF ENDOCRINE-ACTIVE SUBSTANCES (EAS)

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For ecological risk assessment, endocrine disrupters require the establishment of an endocrine mode of action (MoA) with a plausible linkage to a population-relevant adverse effect. Current ecotoxicity test methods mostly incorporate apical endpoints although some also include mechanistic endpoints, at the subcellular through organ level, which can help establish an endocrine MoA. However, the link between these endpoints and adverse population-level effects is often unclear, except for well-documented historical cases (e.g., bird population declines and DDT; dogwhelk population declines and TBT). This presentation will be based upon case studies of endocrine-active substances (EAS) (tributyltin, ethinyl estradiol, perchlorate, trenbolone, propiconazole, and vinclozolin) that were used to evaluate the population relevance of toxicity endpoints in various taxa according to the OECD Conceptual Framework for Testing and Assessment of Endocrine Disrupters in the context of a 2016 SETAC Pellston Workshop™ "*Environmental Hazard and Risk Assessment Approaches for Endocrine-Active Chemicals*." This presentation will focus on examples of potential population relevance from the case study chemicals as well as briefly discuss future needs to better predict population level effects.

A PROPOSED STANDARD NATURAL RESOURCE DAMAGE ASSESSMENT (NRDA) METHOD FOR SMALL TO MEDIUM VOLUME OIL SPILLS

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In Massachusetts, over the last two decades, approximately 70 small to medium volume oil spills less than 10,000 gallons were reported per year. Initiating and completing a natural resource damage assessment (NRDA) for spills of this size can be labor intensive. As such, the adoption of a standard approach for cost effectively assessing natural resource damages resulting from such spills will allow the public to efficiently be compensated for damages and expedite the implementation of compensatory restoration. Consistent with current federal and state NRD assessment

practice, the authors present a method for calculating damages, including interim losses, based on the cost of appropriate restoration actions. The primary factors accounted for in the proposed standard approach include (1) the impacted area, (2) service loss, (3) baseline, (4) recovery, (5) habitat type, (6) discount rate, (7) restoration benefits, (8) restoration costs, (9) resources of special concern, (10) project management costs, and (11) inflation. The proposed method relies on release-specific data historically provided to the Massachusetts Department of Environmental Protection (MassDEP) through the Bureau of Waste Site Cleanup (BWSC) transmittal forms as well as other publicly available data from the Massachusetts Geographic Information System (MassGIS). As such, the proposed standard approach does not create an additional burden by requiring the collection of data that would not otherwise be collected. Additionally, the authors utilize historical data on small to medium volume oil spills to back-test the proposed standard approach and evaluate assumptions.

MEETING A MULTI-DISCIPLINARY MISSION IN A CHANGING CLIMATE USING COASTAL VULNERABILITY INDEXING TO PRIORITIZE PROTECTION OF ECOLOGICAL SERVICES, PUBLIC RECREATION AND CULTURAL AND HISTORIC RESOURCES ON THE TRUSTEES OF RESERVATIONS COASTAL PROPERTIES.

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Thomas O'Shea, Russell Hopping, Trustees of Reservations

Woods Hole Group is working with the Trustees of Reservations (TOR) to conduct a comprehensive assessment of the vulnerability of their coastal properties (35 properties and over 2100 assets) to sea level rise and storm surge inundation. Using a dynamic and probabilistic sea level rise and storm surge model, modelers generate probabilities of inundation in 2030 and 2070. The model provides outputs such as flood probabilities, flood duration, inundation depth and flood pathways. The probabilities are calculated (min, max and spatially weighted average) for each asset on each coastal property. The inundation model outputs are combined with consequence (comparative evaluation of loss of asset due to inundation) data for each asset on each property. Working with Woods Hole Group, the Trustees selected criteria for use in determining the value of each asset. The multiple valuation criteria scores are combined into a consequence score for each asset. A coastal vulnerability index (CVI) is used to strategically rank each asset based on the value and vulnerability. The properties and assets that are both the most valuable to the mission of TOR and the most vulnerable will be at the top of this list. Woods Hole Group identified the most vulnerable properties and asset classes, prepared conceptual adaptation plans for the top 5 vulnerable assets and provided general recommendations for all 35 properties. Currently, Woods Hole Group is developing a more detailed adaptation plan for one of the most vulnerable properties.

Session 5: Toxics Use Reduction

SUSTAINABILITY AND TOXICS USE REDUCTION: CASE STUDIES IN FINDING SOLUTIONS

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Adopted in 1989, the Toxics Use Reduction Act (TURA) is designed to protect public health and the environment while enhancing the competitiveness of Massachusetts businesses. Under the Act, facilities that use large amounts of toxic chemicals are required to report on their chemical use, conduct toxics use reduction planning every two years, and pay a fee. The fees paid by TURA filers support the work of the three TURA implementing agencies:

- The Massachusetts Department of Environmental Protection (MassDEP) is the regulatory body for the program. MassDEP certifies Toxics Use Reduction (TUR) Planners, collects chemical use information and other data submitted by companies, provides compliance guidance, and takes enforcement actions.
- The Office of Technical Assistance and Technology (OTA) is a non-regulatory agency within the Executive Office of Energy and Environmental Affairs. OTA provides free, confidential, onsite technical assistance to Massachusetts manufacturers, businesses, and institutions.
- The Toxics Use Reduction Institute (TURI) provides education and training for companies; sponsors research into the development of cleaner, safer materials and technologies; provides grants to companies, community organizations, and municipalities; convenes business working groups to address specific environmental challenges; conducts policy research and analysis; and provides laboratory and library services.

Recent success stories on finding safer alternatives for communities, small businesses and industry in MA through work at TURI have included:

- Researching alternatives to crumb rubber infills for artificial turf fields. The Institute has compiled information on chemicals, costs, and physical and biological hazards of various field infill materials and made it available to municipalities and other stakeholders involved in decision making in the communities considering artificial fields. Videos about alternatives including TPE and organically managed fields have also been created.
- Providing technical and financial assistance to dry cleaners to convert from solvent use to professional wet cleaning. TURI completed an alternatives assessment to perchloroethylene in dry cleaning and has assisted 16 cleaners in making the transition. Data is collected and analyzed from each cleaner, collectively showing the economic, environmental, health and safety benefits of wet cleaning, all while maintaining top quality in garment care.
- Assisting food processors and beverage manufacturers reduce chemical use. TURI is taking part in a larger project to help the food and beverage sector in MA reduce chemical use, waste, and energy. Specific projects have focused on reducing chemicals used in cleaning and sanitizing processes including at microbreweries, hydroponic farms, and fruit processors. Collaborative research has helped evaluate material choices and potential process modifications.

ACCELERATING THE DESIGN, EVALUATION, ADOPTION, AND SCALE OF SAFER CHEMISTRIES THROUGH SCIENCE, POLICY AND MARKET SHIFTS.

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There are increasing scientific concerns about the health implications of chemicals used in manufacturing processes and products. These concerns are leading to increased consumer, market, and regulatory pressures to eliminate chemicals of concern. Chemical deselection without sufficient consideration of alternatives can lead to regrettable substitutions, where alternatives create new health risks or do not adequately perform. Within this context, the

Chemicals Policy and Science Initiative (CSPI) of the Lowell Center for Sustainable Production at UMASS Lowell is undertaking research and "strategic stakeholder engagement" to drive safer chemistry. We see three important transformations to accelerate safer chemistry: a science transformation (which focuses on enhancing our scientific toolbox to more rapidly evaluate hazards and exposures, compare alternatives, and design new, more sustainable chemicals and materials); a market transformation (increasing both the demand for safer chemicals by chemical users as well as the supply of safer chemistries); a policy transformation (policies that allow rapid preventive actions on problem chemicals and incentivize the design and adoption of safer alternatives).

This presentation will provide an over view of CPSI efforts to accelerate the fields or alternatives assessment and green chemistry. Alternatives assessment has been defined as a "process for identifying and comparing potential chemical and non-chemical alternatives that could replace chemicals or technologies of concern on the basis of their hazards, performance, and economic viability." The science policy field has grown tremendously over the past several years, in part due the publication of the 2015 National Research Council Framework to Guide Selection of Chemical Alternatives. There are a number of gaps in science and practice that an emerging "community of practice" is trying to address.

In many cases, safer alternatives to chemicals of concern may not exist, in which case research and development of green chemistry alternatives is needed. Green chemistry is the design of chemicals and materials that are less hazardous throughout their lifecycles. The Green Chemistry and Commerce Council (GC3), a CSPI initiative, is a 100-member business to business network of companies across sectors and the value chain working collaboratively to advance green chemistry adoption. As with alternatives assessment, there are a number of barriers to implementation that the GC3 attempts to address through collaborative research and dialogue projects. In the end, while alternatives assessment and green chemistry are "solutions-focused," they require traditional "problem-oriented" tools and skillsets of toxicology, exposure assessment, risk assessment and lifecycle assessment. We conclude with some ways in which health and ecosystem scientists can more effectively collaborate with those designing and evaluating safer solutions.

REDUCING RELIANCE ON FLAME RETARDANTS IN GYMNASTICS TRAINING FACILITIES

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Introduction: Gymnasts have been found to have elevated concentrations of brominated and organophosphate flame retardants (FRs) due to their use in gymnastics safety equipment. This finding is consistent with elevated concentrations of FRs measured in gym air and dust, with the highest in and near the loose foam pit. As children may be especially vulnerable, it is precautious to limit exposure to these chemicals when possible. FRs are added to foam pit cubes to meet flammability standards, therefore we aimed to determine fire safety in gyms can be maintained without the use of FRs in the loose foam pit.

Methods: We first quantified the frequency, source, and severity of fires in U.S. gymnastics training facilities by conducting a search of the National Fire Protection Association and FACTIVA databases. Using this information we adapted two representative flammability tests and applied these tests to three pit cube test systems: FR with no cover, FR-free with no cover, FR-free with a polyester cover. Each pit cube system was tested both individually and using a mini test pit. Results were shared with the local Fire Marshal and to inform a Fire Protection Engineer analysis and report for a Massachusetts gymnastics training facility replacing their loose foam pit using FR free foam.

Results: We determined that gym fires are uncommon with less than one identified per year over the past 20 years. The majority of fires occurred at night and tended to be severe. The most common reported source of fire was arson, followed by welding sparks, frayed electrical wire, and cigarette. The flammability testing found that flame retardants

were not needed to resist burning with contact to a smoldering cigarette. The testing also found that the presence of FRs allowed the pit cube to resist igniting when a 1.5 inch flame was applied, however would burn readily in the presence of a 6 inch flame. Under super critical conditions, the presence of FRs in the pit cubes reduced heat release by approximately 4.5 minutes, but had a similar magnitude and duration as when FR-free pit cubes were used. Results were similar for the covered and uncovered FR-free pit cubes. The Fire Marshal determined that FR containing pit cubes were not required in the gym with the presence of a fire detection and suppression system.
Conclusions: Fire behavior of foam pit cubes and pits needs to be considered in the context of overall gym fire safety performance including appropriate design fire, fire detection and suppression, building fire resistance and egress pathways, evacuation plan and training. With these considerations fire safety can be maintained in gyms without the use of FR in the loose foam pit.

BREAKING AWAY FROM OUR TOXIC TRADITIONS: NATURAL DEFENSES FOR HUMAN HEALTH AND AGRICULTURE

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There is little doubt that chemicals have played an enormous role in growing food and protecting against disease over the last century. Pesticides and fertilizers (along with other agricultural practices) have helped farmers ramp up production to feed billions. Before the antibiotic age, infections—from meningitis to strep and staph—reigned as all-too-often-incurable killers. Penicillin saved countless lives, and when it failed another antibiotic took its place. Most of us alive today are beneficiaries of this chemical warfare waged by humans against pest and pathogen. Then came resistance and other unintended side effects, from altered ecosystems to the emergence of opportunistic diseases: from *Clostridium difficile* in humans to *Phytophthora infestans*, once responsible for the Irish potato famine and now increasingly fungicide resistant. fungicides; aggressive weeds crowd out crops; and common pesticides kill off even the beneficial insects. How do we replace the most problematic twentieth-century pesticides, or save our antibiotics so that they are there for us when we most need them? One of the greatest advances over the past century is an improved understanding of ecology – whether a farm field, wetland or our own body. Solutions can now be found in innovative strategies employing technologies that are better informed by ecology from fecal transplants and phage therapy in humans and by using pheromones and bacteria on the farm. Many strategies are borrowed from nature, one of our best allies against these age-old enemies. And they provide us with the opportunity to reduce our dependence on toxic chemicals.

POSTER PRESENTATIONS

XENOESTROGENS AND THEIR INFLUENCE ON THE PROTECTIVE EFFECTS OF PREGNANCY IN THE MOUSE MAMMARY GLAND

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Endocrine disrupting chemicals (EDCs) can mimic the effects of hormones and disrupt development in animals, including during pregnancy and lactation. Pregn
ancy has been shown to have a protective effect on breast cancer, decreasing lifetime risk in females by inducing tumor suppressor pathways. The objective of this study is to determine the effects of two xenoestrogens, oxybenzone (BP-3), found in sunscreen, and propylparaben (PP), found in personal care products, on the protective effects of pregnancy in the mouse mammary gland. Because it can act via estrogen receptor (ER) alpha, we hypothesize that BP-3 will diminish the protective effects of pregnancy; because PP acts via ER beta, we hypothesize that it will support the protective effects of pregnancy. To address our hypothesis, female mice were exposed to either vehicle or one of three doses of each chemical during pregnancy and lactation. A nulliparous group was used that received the vehicle treatment for further comparison. After weaning, involution in the dams was allowed to occur and mammary gland tissue samples were obtained five weeks later. Preliminary analyses indicate that treatment with both BP-3 and PP can alter mammary gland histoarchitecture and change the morphology of the mouse mammary gland after pregnancy. BP-3 induces a mammary gland that more closely resembles a nulliparous one than controls. Studies are in progress to measure RNA and protein levels in the mammary gland using qPCR and immunohistochemical staining. These results suggest that estrogenic EDCs can alter mammary gland development in the mother with effects that are long-lasting.

N-NITROSODIETHYLAMINE (NDEA) EXPOSURES TO ZEBRAFISH (DANIO RERIO) EMBRYOS AND ITS EFFECTS ON ENDOCRINE PANCREAS DEVELOPMENT

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Nitrosamines are chemicals found in charred food and water sources that can cause carcinogenesis and metabolic dysfunction in adults, but little is known about their impact during the gestational period. Nitrosamine exposure during critical windows of embryonic cell differentiation and proliferation may influence metabolic function and disease progression during childhood and adulthood. In this pilot study, transgenic Tg(insulin:GFP) zebrafish
(Danio rerio) were exposed beginning at 3 hours post fertilization (hpf) to the class 2A carcinogen N-nitrosodiethylamine (NDEA) in concentrations of 0, 0.01, 0.1, 1, and 10 µM. At
96 hpf, larvae were imaged in vivo for whole fish morphology, pancreatic β-cell area, swim bladder inflation, and islet deformities. No significant differences were observed across the exposure groups for fish length and β-cell area. In the 10 µM exposure group, yolk sac area was significantly reduced, and the proportion of swim bladder inflation was significantly increased as compared to controls. In the 0.1 µM exposure group, significantly more islet deformities occurred as compared to the control group, clustering in hollow, fragmented, and posteriorly placed shapes. While the results of this study did not reach significance for all exposure groups, it appears that NDEA mimics the effects of other endocrine-disrupting chemicals, demonstrating non-monotonic dose-response curves. These results suggest that the embryonic endocrine pancreas may be sensitive to nitrosamine exposures, and that these exposures could result in congenital pancreas defects associated with susceptibility to diabetes and metabolic dysfunction.

PANCREAS DEVELOPMENT AND NUTRIENT UPTAKE AND UTILIZATION ARE DISRUPTED BY EMBRYONIC EXPOSURES TO THE ENVIRONMENTAL TOXICANT PERFLUOROOCTANESULFONIC ACID IN THE ZEBRAFISH

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Several persistent and ubiquitous environmental toxicants have been categorized as diabetogens or obesogens due to their numerous associations with these conditions in adult humans. These compounds often produce deleterious effects in the pancreas, which secretes glucoregulatory hormones and proteases and lipases essential for digestion. Compounds such as the surfactant perfluorooctanesulfonic acid (PFOS) have been detected in biological samples from nearly 100% of the population, and are associated with increased risk for diabetes in adults. However, the developmental consequences of these exposures which may contribute to these adult phenotypes require elucidation. We hypothesized that embryonic exposures to PFOS would alter pancreas development and disrupt embryonic nutrition processes associated with pancreas function. Zebrafish embryos were exposed to 16 or 32 µM PFOS or 0.01% DMSO (control) throughout embryonic development (3-96 hpf). Transgenic embryos from *Tg(insulin:GFP)* and *Tg(ptf1a:GFP)* strains were imaged *in vivo* at 96 hpf to visualize endocrine and exocrine pancreatic development, respectively. Larval morphometry was examined to probe yolk utilization and fish growth. RNA was isolated to examine expression of genes central to pancreas function, and biochemical assays were performed to quantify total embryonic nutrient concentrations. Beta cell mass and insulin gene expression were both decreased concordantly with increasing PFOS exposure at 96 hpf. Total pancreas length and expression of protease genes were also decreased by PFOS exposure. The rate of yolk utilization was increased by 6-10% due to 16 and 32 µM PFOS, respectively, suggesting increased embryonic uptake of total yolk nutrients. However, total embryonic concentrations of the bulk yolk nutrients cholesterol, triglycerides, and glucose were decreased with increasing PFOS concentrations. These PFOS-exposed larvae were also shorter than control fish at 7 dpf. Free fatty acid profiles also differed between control and PFOS-exposed embryos. Total embryonic concentrations of several saturated free fatty acids such as stearic acid were increased by treatment, while concentrations of beneficial unsaturated free fatty acids such as α-linolenic acid were decreased by treatment. Overall, this work demonstrates that embryonic toxicant exposures can perturb pancreas development, and can alter the uptake and processing of yolk nutrients during development.

EMBRYONIC EXPOSURE TO MONO(2-ETHYLHEXYL) PHTHALATE (MEHP) ALTERS PANCREATIC ORGANOGENESIS IN ZEBRAFISH *(DANIO RERIO)*

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Mono(2-ethylhexyl) phthalate (MEHP) is the bioactive metabolite of di(2-ethylhexyl) phthalate, a plasticizing agent and persistent environmental contaminant associated with obesity, developmental abnormalities, and oxidative stress. We have previously identified the pancreas as a sensitive target of oxidative stress during embryonic development. Our objective was to determine whether MEHP exposure during organogenesis affects pancreatic development. Zebrafish embryos were exposed to 0 or 200 µg/L MEHP at 3 hours post fertilization (hpf) through 96 hpf. Fluorescence microscopy of *Tg(ins:GFP)* zebrafish determined that MEHP exposure significantly decreased ß-cell area at all timepoints (48, 72, 96, 168 hpf). *Tg(gcga:GFP)* embryos exposed to MEHP showed a similar decrease in α-cell area in the islet across the same time points. *Tg(ptf1a:GFP)* embryos were assessed at 80 and 168 hpf for exocrine pancreas length. MEHP exposure decreased growth of the exocrine pancreas tail resulting in shortened ratios of pancreas to fish length. All deviant pancreas morphologies occurred in the absence of any gross morphological malformations. Expression of pancreas hormone genes *insa* and *gcga* was significantly reduced by MEHP exposure compared to controls at 96 hpf as demonstrated by RT-PCR. These data indicate that embryonic exposure to MEHP can negatively impact pancreatic organogenesis. This work was supported by a grant from the NIH (R01ES025748).

INVESTIGATION OF PESTICIDES PENETRATION AND PERSISTENCE ON HARVESTED AND LIVE BASIL LEAVES USING SURFACE-ENHANCED RAMAN SCATTERING MAPPING

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Understanding pesticide behavior in plants is significant for effectively applying pesticides and in reducing pesticide exposures from the plant. This study aimed to investigate the penetration and persistence of pesticides applied on harvested and live basil leaves. Surface-enhanced Raman scattering (SERS) mapping was applied for *in situ* and real-time tracking of pesticides over time using penetrable gold nanoparticles as probes. The results showed that both the systemic pesticide thiabendazole and the non-systemic pesticide ferbam penetrated more rapidly and deeply into the live leaves than the harvested leaves after surface exposure of 30 min to 48 h. The effects of leaf integrity and size on thiabendazole penetration were also evaluated on live basil leaves. Thiabendazole when applied onto intact leaves penetrated deeper than when applied onto damaged leaves. Older leaves with a mass of 0.204 ± 0.019 g per leaf (45 days after leaf out) allowed more rapid and deeper penetration of pesticides than when younger leaves with a mass of 0.053 ± 0.007 g per leaf (15 days after leaf out) were used. The degradation of thiabendazole on live leaves was detected after 1 week whereas the apparent degradation of ferbam was detected after 2 weeks. In addition, the removal of pesticides from basil was more efficient when compared with other fresh produce possibly due to the specific gland structure of basil leaves. The information obtained here provides a better understanding of the behavior and biological fate of pesticides on plants.

HEALTHY FISH, HEALTHY PEOPLE: USING FISH TO UNDERSTAND ECOLOGICAL AND HUMAN HEALTH IMPACTS OF EARLY LIFE EXPOSURES TO SUPERFUND CHEMICALS ON METABOLIC AND BONE DEVELOPMENT.

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Fish inhabiting the New Bedford Harbor (NBH), Massachusetts, marine Superfund site can serve both as biological models for contaminant effects and indicators of human dietary exposure, contributing importantly to the assessment of ecological and human health risks of contaminant exposure. Polychlorinated biphenyls (PCB) and tributyltin (TBT) are bioaccumulative contaminants associated with NBH industries that belong to a growing class of metabolism-disrupting compounds believed to contribute to obesity, liver steatosis, and Type 2 diabetes in humans and other species. Here we show that embryonic exposure to PCBs and TBT produced phenotypic abnormalities and altered the expression of genes related to metabolic homeostasis in laboratory-reared killifish (*Fundulus heteroclitus*), an ecologically-important NBH fish. These biological effects suggest perturbations to metabolic and bone homeostasis in fish, consistent with effects seen in mammalian species; future transcriptomic analyses will provide insight into the underlying molecular mechanisms of toxicity for these compounds in fish. In a complementary investigation, we also used information from NBH seafood as a proxy for human dietary exposure to harbor-based Superfund chemicals, and show here that PCBs in human-consumed species from NBH have generally declined since 2003. This information may be useful in understanding the contribution of chemical metabolic disruptors in human obesity and metabolic disease. The combination of mechanistic studies using fish and the assessment of potential human exposure through consumption of contaminated seafood provides an effective and holistic approach to characterize both ecological and human health risks of exposure to environmental chemicals, including those frequently found at sites highly contaminated with multiple Superfund chemicals.

COMPARATIVE CYTOTOXICITY OF CHLORINATED AND BROMINATED HALOBENZOQUINONES ON HUMAN COLON EPITHELIAL CELLS

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Disinfection, an essential step in providing safe drinking water to public, results in undesirable formation of disinfection by-products (DBPs) that have been associated with health concerns. DBPs such as trihalomethanes and haloacetic acids have been regulated by the US Environmental Protection Agency. A group of unregulated DBPs, halobenzoquinones (HBQs), have been associated with increased risk of bladder and colon cancers. High occurrence frequencies of 2,6-dichloro-1,4-benzoquinone and 2,6-dibromo-1,4-benzoquinone have been reported in effluent samples from US and Canadian utilities. However, the exposure to these HBQs in the distribution systems is governed by their stability under typical drinking water conditions. Varying pHs and the presence of disinfectant residual can result in transformation of HBQs into other products that might also be of health concern. Therefore, a comparative toxicity analysis was performed by exposing immortalized normal human colon epithelial cells (CCD 841 CoN) to HBQs and their transformation products. This study has implications with respect to health and regulations as well as drinking water treatment strategies aimed at minimizing the formation of and exposure to unregulated DBPs.

PYRETHROIDS INCREASE TETRODOTOXIN-SENSITIVE SODIUM CURRENTS EXPRESSED IN RAT BRAIN TISSUE MICROTRANSPLANTED INTO XENOPUS LAEVIS OOCYTES.

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Microtransplantation of mammalian neurolemma is a tool to study the structure and function of transmitter receptors and ion channels associated with the central nervous system. *Xenopus* oocytes microtransplanted with rat brain tissue from postnatal day 15 and 90 rats were used to examine the effects of deltamethrin and permethrin on native voltage-sensitive sodium channels. Automated western blot analysis indicated that Nav1.2 was the predominant isoform expressed in post natal day (PND) 15 and PND 90 rats. Plasma membranes prepared from microtransplanted oocytes confirmed that these rat channels were successfully microtransplanted to the membranes of oocytes. Using a high throughput two electrode voltage clamp (TEVC) electrophysiological system of microtransplanted oocytes, we were able to detect currents that were sensitive to tetrodotoxin (TTX), □-conotoxin MVIIC, tetraethylammonium (TEA) and niflumic acid (NFA) indicating the presence of multiple voltage-sensitive ion channels (voltage-sensitive sodium, calcium and potassium channels, respectively) and calcium-activated chloride channels. In this study, we compared the effects of a classic Type I pyrethroid (permethrin) to a Type II pyrethroid (deltamethrin) on native VSSC currents in PND 15) and PND 90). Both deltamethrin and permethrin increase the inward TTX-sensitive voltage-sensitive sodium channel currents pharmacologically isolated in the presence of NFA in a concentration-dependent manner. These results indicate that this method is amenable to studying environmental contaminants that target mammalian neuronal ion channels and will also have utility for the study of additional agents shown to cause acute, chronic and developmental neurotoxicity in other mammalian neuronal tissues, including those from knockout mice models and humans.

PROBABILISTIC ECOLOGICAL RISK ASSESSMENTS – ARE THEY WORTH THE TIME?

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Ecological risk assessments evaluate risks to receptors exposed to media potentially impacted by site activities. Typically, a deterministic ecological risk assessment (DERA) is conducted for representative receptors with upper-end exposure assumptions (*e.g.*, 100% diet, home ranges equal to site) and central tendency values (*e.g.*, body weight) that ignore the variability of the available data. In a DERA, the inputs for the exposure models and effects

concentrations are single values that are used to calculate a simple ratio of effect (*i.e.*, hazard quotient [HQ]). Alternatively, probabilistic ecological risk assessments (PERA) reduce the uncertainties associated with limited data and modeling variability by characterizing risks based on the magnitude of risk as well as the probability of occurrence.

We conducted an ecological risk assessment for six terrestrial receptors exposed to metals in surface soils, surface water, and associated dietary items using both deterministic and probabilistic methods. The two methods differed in exposure parameters (*i.e.*, body weight, home range, percentage of diet). For the PERA, we also compared two methods of calculating ingestion rates (diet-specific field metabolic rates [FMR] *vs.* FMRs for representative animal groups). To simplify the process, the same exposure point concentrations (EPCs) and toxicity reference values were used. We compared DERA HQs to PERA HQ ranges and determined which input parameters contributed the most to the overall risk results.

We determined that in addition to producing more realistic mean HQ values compared to the DERAs, the PERAs can also provide more conservative estimations of risk for receptors with large home ranges. For receptors with small home ranges, the percentage of terrestrial invertebrates in the diet impacted the PERA mean HQs by up to a factor of 4. When comparing PERA HQs using the two different ingestion rates, risks using the diet-specific FMRs resulted in lower HQs, but with more variability than HQs using FMRs for representative animal groups. While performing PERAs requires more time, PERA HQs provide information on the magnitude of risk and the probability of occurrence. DERA HQ values only note that the values are conservative, but provide no information on how conservative they are. Consequently, information regarding the probability of occurrence could vastly improve remediation goals and decisions.