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Anaheim, California March 15–19, 2020

Important Dates and Deadlines:

- **May 15, 2019:** Scientific Session/CE Course Proposal Submission Deadline
- **October 9, 2019:** SOT Awards Nomination and Application Deadline for Most Awards
- **October 18, 2019:** 2019 Abstract Submission Deadline
- **October 18, 2019:** Undergraduate Diversity Award, Perry J. Gehring Diversity Student Travel Award, and Pfizer SOT Undergraduate Award Application Deadline

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suggest that treatment with citronellal (over this dose range and duration) did not result in large changes in peripheral nerve, motor, or somatosensory function. These results do not preclude anti-nociceptive properties after acute treatments. *This is an abstract of a proposed presentation and does not necessarily reflect US EPA policy.*

1332 Propranolol as a Novel Treatment for Gulf War Illness in a Preclinical Mouse Model

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Gulf War Illness (GWI) is a multi-symptom, neuroimmune-based disorder that presents with features similar to sickness behavior. Unfortunately, current treatments for GWI tend to focus on managing symptoms as opposed to addressing the underlying cause of the illness. Using a preclinical mouse model, we have found that GWI is associated with an exacerbated neuroinflammatory response to immune challenge, like lipopolysaccharide (LPS) exposure, and the activation of microglia. Interestingly, β-adrenergic antagonism has been found to inhibit microglial activation and its associated release of inflammatory cytokines. Here, we tested the therapeutic potential of the beta-blocker propranolol in our established mouse model of GWI. In this model, mice are exposed to the stress hormone corticosterone (CORT; 200 mg/L) in the drinking water for 7 days followed by a single injection of diisopropyl fluorophosphate (DFP; 4 mg/kg, i.p.) to model the "in theater" conditions of high physiological stress and potential nerve agent exposure. This is then followed by periodic administration of CORT for 7 days every other week to a total of 5 weeks with a systemic LPS challenge (0.5 mg/kg, s.c.) on the final day. Propranolol (20 mg/kg, i.p.) was given during or outside of CORT exposure. Mice were sacrificed 6 hours after LPS challenge and brain cytokine mRNA expression was evaluated by qPCR. We found that propranolol significantly reduced the neuroinflammation instigated by the GWI exposure model when given during CORT exposure. In particular, treatment reduced cytokine expression in the GWI exposure group to levels comparable to CORT+LPS, which models a normal response to inflammatory challenge. These initial studies indicate the potential for propranolol to treat the underlying neuroinflammation associated with GWI and to return veterans to a healthy neuroimmune functional state. *Disclaimer: The findings and conclusions in this report are those of the authors and do not necessarily represent the official position of the National Institute for Occupational Safety and Health, Centers for Disease Control and Prevention.*

1333 Functional Characterization of Neural Network Activity in Human iPSC-Derived Neuron/Glial Co-Cultures

T. Molla, L. Saavedra, P. Zhou, J. Hsieh, A. Fua, H. Liu, J. Davila, and D. Haag. *NeuCyte, Sunnyvale, CA.* Sponsor: I. Pessah

The increasing amount of chemicals with human exposure, and the accumulating evidence for interspecies differences regarding adverse effects on the CNS, call for scalable *in vitro* neurotoxicity screening tools using human cells. Generating neuronal cells through differentiation from induced pluripotent stem cells (iPSCs) carries a great potential to overcome the inaccessibility of human primary tissue and accelerate cell-based assays. In combination with multi-electrode array (MEA) readouts, neural activity in response to chemical compounds can be quantified allowing neurotoxicity screening. However, the assessment of neuroactive effects in human-derived cell-based assays remains challenging due to cell type variability and poorly defined baseline physiology. Here, we describe a new screening platform using highly functional neural cultures with defined cell ratios consisting of excitatory and inhibitory neurons that were separately generated by direct conversion from human iPSCs (NeuCyte SynFire™), as well as primary human astroglial cells. The reduced complexity of this iPSC-derived co-culture system, compared to primary rodent cultures, enables a detailed molecular and functional characterization to define its applicability domain and develop screening assays. Therefore, we conducted comprehensive transcriptome profiling at different maturation time points of the co-cultures and tested altered neuronal firing, bursting and synchrony metrics in response to a set of 15 agonistic and 15 antagonistic tool compounds targeting neuronal signaling (e.g. $GABA_AR$, AMPAR, NMDAR, AChR, D1/2R, and 5-HTR) on MEAs. We then correlated dose-dependent responses with expression patterns at the different co-culture time points to determine sensitivity, specific neuroactivity profiles and potential assay windows for interference with these pathways. Furthermore, we confirmed specific responses by patch clamping of matched neuronal/glial co-cultures. This yielded a detailed pharmacological characterization of NeuCyte's human iPSC-derived neuron/glial co-cultures as baseline and reference for multiple neurotoxicity testing applications.

1334 Nanomolar Tetrabromopyrrole Alters Ca^{2+} Dynamics in Cortical Neuronal Networks by Selective Modification of Ryanodine Receptors and Micromolar Is Neurotoxic Due to SERCA Pump Inhibition

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Naturally synthesized marine organohalogens (MOH) and their anthropogenic homologs produced as disinfection byproducts (DBP) are an emerging environmental health concern because several have been identified to exhibit potent biological activities in model systems, including cytotoxicity, genotoxicity, carcinogenicity and developmental toxicity. The molecular mechanisms mediating toxicity are poorly understood. Recently we discovered that several specific MOH and DBP measured in environmental and biological samples, including halopyrroles, halobipyrroles, haloindoles, and hydroxylated polybrominated diphenylethers directly modify ryanodine receptors and SERCA pump activity, two key proteins anchored within sarcoplasmic/endoplasmic reticulum (SR/ER) that work in physiological opposition to tightly regulate net ER/SR Ca^{2+} dynamics and thereby shape meaningful Ca^{2+}-dependent cellular processes. Using intact HEK293 cells null for ryanodine receptors (RyRs) expression and those that stably express RyR1, we demonstrate that tetrabromopyrrole (TBP) selectively sensitizes RyR1 channels to caffeine-triggered Ca^{2+} release only in RyR1-expressing cells. TBP at higher concentrations also depletes of SR/ER Ca^{2+} stores in both null and RyR1 expressing cells commensurate with its lower potency to inhibitory SERCA in biochemical assays. Exposure of primary neuronal/glial co-cultures derived from newborn mice shows that TBP inhibits the frequency and amplitude of spontaneous Ca^{2+} oscillations (IC_{50}=246 and 426nM, respectively), whereas >1μM produces a sustained rise in cytoplasmic Ca^{2+}. Subchronic (24HR) exposure to TBP caused loss of neuronal/glial viability using the MTT assay (EC_{50}=12.4μM). These results show that nM TBP selectively targets RyR-mediated Ca^{2+} dynamics in a manner that has been shown to affect neurodevelopment, whereas low-μM exposures causes overt neurotoxicity, likely mediated by the combination of RyR activation and SERCA inhibition. *Supported by NIEHS grants ES030318 and ES014901.*

1335 Tetrabromobisphenol A Alters ABC Transport at the Blood-Brain Barrier

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Tetrabromobisphenol A (TBBPA, CAS No. 79-94-7) is a brominated flame retardant (BFR) used in 90% of epoxy coated circuit boards. Exposures to TBBPA can disrupt mitogen-activated protein kinase (MAPK), estrogen, thyroid and peroxisome proliferator-activated receptor (PPAR) signaling pathways. Since these pathways also regulate transporters of the CNS barriers, we sought to determine the effect of TBBPA on the expression and activity of three major ABC efflux transporters of the blood-brain barrier (BBB). Using a confocal based assay, we measured the *ex vivo* and *in vivo* effects of TBBPA on P-glycoprotein (P-gp), Breast Cancer Resistance Protein (BCRP), and Multi-Drug Resistance Protein 2 (MRP2) transport activity in rat brain capillaries. We found TBBPA (1-1000 nM) had no significant effect on MRP2 transport in either sex. However, low concentrations of TBBPA (1-100 nM) significantly decreased BCRP transport activity in both sexes. Additionally, TBBPA exposures (1-100 nM), elicited a sex-dependent response in P-gp transport: increasing transport in males and decreasing transport in females. All TBBPA changes in transport were measured within a TBBPA dose and time of 1-1000 nM and 1-6 hours respectively. Inhibitors of either transcription or translation abolished the TBBPA dependent increases in male P-gp transport. Western blot and immuno-fluorescent assays confirmed the TBBPA dependent P-gp expression increases in males and decreases in females. Inhibition of PPAR-γ by GW9662 abolished the TBBPA dependent increases in males but not the decreases in females. However, the TBBPA decreases in female P-gp transport were blocked by the ER-α antagonist, ICI. This work indicates that environmentally relevant concentrations of TBPPA (1-100 nM) alter ABC transporter function at the blood-brain barrier. Moreover, changes in BBB permeability alters brain homeostasis, modifies CNS drug delivery and increases brain exposure to harmful xenobiotic toxicants. *This work was supported by the Intramural Research Program at NCI/NIEHS, ZIA BC 011476.*