to nanoaerosols from M4 weapon fire reduced cell viability greater than from the M9. Our novel exposure device allows for realistic respiratory exposure by depositing NMs on eight separate *in vitro* lung cell cultures at the air-liquid interface. Distribution A: Approved for public release; distribution unlimited. *(PA Case No. 88ABW-2016-4456, Date 15 Sep 2016)*

S 2287 Big Data, Meet Chemical Carcinogenesis! Are There New Solutions for an Old Problem?

U. Apte. *University of Kansas Medical Center, Kansas City, KS.*

The ability of chemicals to cause cancer is a highly relevant end point with a significant public health impact. Determining the mechanisms by which chemicals modulate normal cellular pathways to induce neoplastic transformation and growth is central to the discipline of toxicology. New data obtained via development of new animal models combined with high throughput 'omics technologies has revolutionized the way we identify modes of action (MOA). These new "Big Data"-assisted technologies have highlighted several novel molecular targets involved in chemical carcinogenesis and changed our understanding of MOA. Development of molecular networking tools has revealed novel gene networks that regulate neoplastic growth secondary to chemical exposure. A completely unbiased adverse outcome pathway analysis is yet another approach that can revolutionize understanding the mechanisms of chemical carcinogenesis. Despite several key advances, major challenges remain in employing the new systems biology based big data techniques to chemical carcinogenesis research. This symposium will bring together scientists bringing "Big Data" methodologies to the chemical carcinogenesis field. The new frontiers of using data analytics to study chemical carcinogenesis and the challenges ahead will be discussed.

S 2288 Mode of Action in Chemical Carcinogens: Setting the Baseline

J. Klaunig. *Indiana University, Bloomington, IN.*

Understanding the mode of action of chemical carcinogens is critical for risk assessment of the chemicals. Every decade since the 1940s has seen major advances in our understanding of the mechanism by which chemical induce cancer. It is now apparent that the previous simplistic view that chemicals that induce cancer interact with DNA, induce a mutation, which results in the formation of a neoplasm, is not complete. The advent of "Big Data" technologies, including global gene expression analyses, genome wide association studies, and deep sequencing techniques, which reveal disease specific posttranscriptional changes, has revolutionized our understanding of pathophysiology. As our knowledge base increases, it is evident that modifications of multiple intracellular and extracellular processes by chemical agents contribute to the induction of a neoplasm. Chemical modulation of metabolism, gene expression, DNA repair processes, immune surveillance, inflammation, cell to cell communication, and changes in target cell function and structure contribute to the formation and growth of preneoplastic cells and their progression to the malignant state. The multitude of changes in the target cell and its microenvironment must be considered in applying mode of action analysis to potential carcinogenic human risk. Whereas the lessons learned from "Big Data" analyses are extremely valuable, they come with caveats, which should be critically considered when using these data. Using our knowledge of liver cancer, this talk will discuss the accepted theories of chemical carcinogenesis, and expand upon boons and bans of using "Big Data" to further our understanding of chemical carcinogenesis mechanisms.

S 2289 Integrating Toxicogenomics Data into Adverse Outcome Pathways for Cancer

C. Corton. *US EPA, Research Triangle Park, NC.*

As the toxicology field continues to move towards a new paradigm in toxicity testing and safety assessment, there is the expectation that models will be increasingly developed and refined to enable better prediction of cancer risk from short-term tests in animals or even cultured cells. The growing number of publically available genomic profiling studies from chemically-treated animals and cells provides opportunities to construct models for prediction of molecular initiating events (MIEs) and key events (KEs), and assess linkages between the events as KE relationships (KERs) in cancer Adverse Outcome Pathways (AOPs). Using a number of computational approaches, including the comparison of wild-type and nullizygous mice, as well as annotated conditions in which key events in known cancer pathways occur, we derived gene expression biomarkers that accurately predict events, including activation of transcription factors (TFs) (e.g., aryl hydrocarbon receptor (AhR), constitutive activated receptor (CAR), peroxisome proliferator-activated receptor alpha (PPARalpha), and sex steroid receptors) and increases in cytotoxicity and downstream KEs including increases in oxidative stress (using Nrf2 activation as a surrogate), and inflammation. These biomarkers were used to comprehensively assess MIE/KE modulation by ~200 chemicals linked to known incidences of hepatocellular adenomas and carcinomas in rodents under chronic exposure conditions. The analysis highlighted a number of features of chemically-induced mouse liver tumor induction: 1) most carcinogenic chemicals activated multiple MIEs; 2) simultaneous assessment of known AOPs uncovered unexpected KERs between TFs (e.g., activation of CAR and Nrf2); and 3) *in vitro* models evaluated were generally poor surrogates of the intact liver in prediction of events. Because chemical-independent (and potential modulating) factors that regulate the MIEs/KEs are simultaneously evaluated in the database (e.g., diet, genetic background, and life stage), the integrated predictions may inform assessment of cumulative risk using AOPs. (This presentation does not represent EPA policy.)

S 2290 Warburg-Like Differential Gene Expression and Metabolic Reprogramming during Progression of Cancer Pathogenesis: Lesson Learned from Liver Cancer Studies

T. Zacharewski. *Michigan State University, East Lansing, MI.*

2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) and related compounds elicits dose-dependent hepatic fat accumulation, inflammation, and fibrosis in mice that may predispose animals to the development of hepatocellular carcinomas. To further investigate the mechanisms involved, we have manually and computationally integrated complementary ChIP-Seq, RNA-Seq and metabolomic data sets to identify alterations in central carbon and amino acid metabolism consistent with these TCDD-elicited phenotypic responses. More specifically, AhR ligands induced a Warburg-like effect that includes glycolytic reprogramming as well as increased glutaminolysis, NADPH production, and ascorbic acid synthesis, in support of ROS defenses and excessive extracellular matrix deposition. The roles of iron overloading and heme production in the progression of steatosis to steatohepatitis with fibrosis will also be discussed. This presentation will demonstrate the integration and interpretation of 'omic data to identify gene expression and metabolite changes within specific converging metabolic pathways involved in the progression of AhR-mediated metabolic disruption leading to heptocellular carcinoma.

S 2291 NGS Reveals Novel Targets and MOA in Chemical Carcinogenesis

U. Apte. *University of Kansas Medical Center, Kansas City, KS.*

Nuclear receptors are central to pathophysiology and are often involved in the mechanisms of toxicity induced by chemicals. Large majority of chemical carcinogen classified as tumor promoters act via activating nuclear receptor(s). The role of certain nuclear receptors such as PPARa, and CAR, has been well recognized in chemical carcinogenesis. Recent studies indicate that other nuclear receptors previously not associated with cancer pathogenesis play critical role in pathogenesis of cancer, especially chemical carcinogenesis. This presentation will highlight the previously unknown role of nuclear receptors such as HNF4a in chemical carcinogenesis. Whereas the role of these receptors in metabolic processes and differentiation has been known, new studies indicate these proteins are central in cancer pathogenesis either via their canonical role or via non-canonical actions driven by chemical exposure. The role of these receptors as novel targets of chemical carcinogens will be discussed and the use of next generation sequencing (NGS) data in better understanding MOA involving HNF4a in chemical carcinogenesis by chemicals such as PFOA and endobiotics such as bile acids will be highlighted.

S 2292 Chemically-Induced Neuroinflammation and "Sickness Behavior" Disorders

J. O'Callaghan. *CDC-NIOSH, Morgantown, WV.*

Neuroinflammation is a dominant theme in contemporary neuroscience. This is not surprising given the number of neurological disease states, e.g. Alzheimer's Disease, Parkinson's Disease, Huntington's Disease, where neuroinflammation has been implicated. Thus, a clear association has emerged among neurodegenerative disorders, and the elaboration of proinflammatory cytokines and chemokines in the CNS, the core feature of the neuroinflammatory condition. While neuroinflammation often occurs in association with damage to neurons and glia, it

also can occur in the absence of neurodegeneration, e.g. where elevated concentrations of proinflammatory cytokines are seen with systemic infection. In these circumstances, neuroinflammation is associated with sickness behavior, i.e. a constellation of symptoms manifested in loss of appetite, fever, muscle pain, fatigue, and cognitive problems. Typically, sickness behavior accompanies an inflammatory response that resolves with time, with gradual restoration to homeostasis. However, chronic sickness behavior syndromes can also occur, and may be instigated or exacerbated by chemical exposures, both from the environment and pharmaceuticals. In this symposium, we bring together two junior investigators and two senior investigators to provide an overview of the central/peripheral immune system interactions that can contribute to the sickness behavior condition, and present recent preclinical and clinical data, as well as data from experimental models, on three sickness behavior disorders: Chemobrain, Gulf War Illness, and Chronic Fatigue. These presentations serve as examples of the chronic neuroinflammatory condition as an important chemical exposure issue, with implications beyond the disorders and exposures to be presented. The goal of the symposium is to provide a framework for considering alterations in the normal inflammatory response of the nervous system as a basis for complex neurological and physiological disorders that can be initiated or exacerbated by chemical or pharmaceutical exposure.

S 2293 Overview of Neuroinflammation and Sickness Behavior

C. A. McPherson. *NIEHS/NIH, Research Triangle Park, NC.* Sponsor: J. O'Callaghan.

Neuroinflammation in the central nervous system (CNS) is mediated in part by the production of pro- and anti-inflammatory cytokines that initiate signaling contributing to injury resolution and repair, facilitating a return of CNS homeostasis. This process can occur in the presence or absence of structural neuropathology. One clinical manifestation of neuroinflammation is the induction of sickness behaviors including malaise, lethargy, and hypopagia. This coordinated response of sickness behaviors is driven by pro-inflammatory cytokines, serving as an acute phase response to infection and injury that enhances survival advantage to the host. However, if unresolved, they may develop into an adverse state that has the potential to develop into a chronic condition. Types of sickness behavior that manifests as chronic fatigue, sick building syndrome, cognitive deficits following chemotherapy, and persistent effects following chemical exposure under stressful conditions, remain of concern for underlying etiology and therapeutic intervention. We now know that a line of communication exists between the peripheral and the central immune systems that may offer an avenue to understand the etiology of such disease states. Peripheral inflammation can be communicated to the CNS via circulating monocytes, perivasculature microglia, endothelial cells, or circumventricular organs. Contribution of this communication in an inflammatory response in the CNS has been demonstrated in both rodent and human studies. Additionally, recent studies have demonstrated a lymphatic drainage system for the CNS in rodents that may allow communication to the peripheral immune system. This cross-talk between the periphery and CNS may result in the propagation of inflammation in the brain vasculature and the brain, including but not limited to, expression of the pro-inflammatory cytokines interleukin (IL)-1, IL-6, tumor necrosis factor (TNF)a. The interplay between peripheral and central inflammation may allow for a biofeedback system to maintain a chronic peripheral/central inflammatory condition, and underlie diverse neuropathology or manifestation of sickness behavior resulting from various chemical exposures or CNS disorders..

S 2294 Chemobrain

C. J. Heijnen. *University of Texas M.D. Anderson Cancer Center, Houston, TX.* Sponsor: J. O'Callaghan.

Cognitive impairment following chemotherapy, termed "chemobrain," is a relatively common side effect, affecting approximately 25% of patients receiving the treatment. While many cases of chemobrain are temporary, cognitive deficits may persist for many years after the treatment. As with other "sickness behavior" disorders, neuroinflammation and associated mitochondrial dysfunction are thought to contribute to the chemobrain condition. Cisplatin is one of the chemotherapeutics shown to induce persistent cognitive deficits representative of chronic "sickness behavior." In an experimental model, C57Bl/6J mice received two cycles of cisplatin treatment (2.3 mg/kg/cycle), and upon subsequent testing in the Novel Object Recognition test and Y-maze showed impaired learning and memory. This deficit was accompanied a deficit in mitochondrial respiratory function as evidenced by a marked decrease in spare respiratory capacity of synaptosomes. This was also accompanied by damage to the myelin sheath as indicated by immunostaining of myelin basic protein, and a decrease in number of doublecortin+ new neurons within the subventricular zone (SVZ) and the dentate granule cell layer of the hippocampus, each of which has been associated with conditions of chronic neuroinflammation. Nasal administration of mesenchymal stem cells (MSC) in mice following chemotherapy treatment mitigated cisplatin-induced cognitive impairment, synaptosome mitochondrial deficit, and histological changes in myelin and newly generated neurons in the SVZ and SGZ. Despite the prevalence of chemobrain among patients receiving chemotherapeutics, no reliable treatments for the cognitive deficits have been identified. Our data suggest that treatment with MSC may represent a realistic therapeutic strategy for the prevention of chemotherapy-induced cognitive impairment and underlying neuroinflammation.

S 2295 Gulf War Illness

L. T. Michalovicz. *CDC-NIOSH, Morgantown, WV.*

Gulf War Illness (GWI) is a chronic multi-symptom disorder characterized by persistent headaches, muscle pain, chronic fatigue, memory loss, confusion, gastrointestinal disturbances, and rashes. While the illness is believed to be the result of various toxic exposures during the 1991 Persian Gulf War, the underlying biological cause of this persistent illness is still being investigated. The alignment of GWI with sickness behavior, a collective of behavioral symptoms that has been associated with neuroinflammation, suggests that GWI may also be the result of a persistent inflammatory state in the brain. By evaluating animal models subjected to similar "in theater" exposure conditions, it has become apparent that exposures to toxicants like the nerve agent, sarin, or its surrogate, diisopropyl fluorophosphate (DFP), and various organophosphate insecticides, e.g., chlorpyrifos, cause significant neuroinflammatory effects. When these exposures are coupled with a stressor or a stressor mimic, such as exogenous corticosterone, the resultant neuroinflammation, as indicated by enhanced mRNA expression of broad categories of cytokines and chemokines assessed by qPCR, is exacerbated and maintained over a longer duration. Building upon these observations, while a single exposure scenario may not alter the baseline response threshold, a combination of stress and chemical toxicants (e.g., under wartime conditions) can be observed to shift this threshold, priming the brain in a manner that will result in an exacerbated response to a subsequent inflammatory challenge. We show that this priming shifts the susceptibility of the CNS to immune challenges, resulting in the potential for an exacerbated sickness behavior response to even mild stimuli, such as a low dose of the bacterial mimic, lipopolysaccharide (LPS).

S 2296 Chronic Fatigue

N. G. Klimas. *Nova Southeastern University, Fort Lauderdale, FL.* Sponsor: J. O'Callaghan.

Chronic fatigue (CF) is characterized by fatigue that persists for six months or more. CF, in combination with a minimum of 4 of 8 symptoms, including memory impairment, pain, headaches, and sleep disturbance, and the absence of diseases that could explain these symptoms, constitute the case definition for chronic fatigue syndrome/myalgic encephalomyelitis (CFS/ME). This pattern of etiology has been attributed to a number of factors, including chemical exposures. As with other "sickness behavior" disorders, neuroinflammatory mediators are thought to underlie CF. Recent advances in the understanding of CF pathogenesis has led to the elucidation of patterns of CF biomarker expression, providing the first hints of a molecular signature of the disorder, including factors associated with inflammation, immune system activation, autonomic dysfunction, neuroendocrine, and altered functioning in the hypothalamic-pituitary-adrenal axis. Further in depth systems biology analysis has identified several cytokines, e.g. IL-1β, IL-6, and IL-8, that may function as biomarkers CFS/ME, and may provide an indication of the duration and severity of illness. Moving forward, the use of these biomarkers will provide an opportunity to understand potential exposures/triggers that result in CF and to develop effective treatments.


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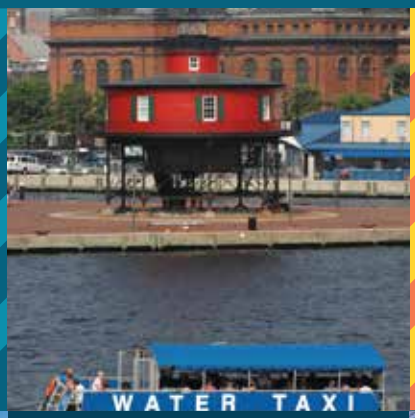

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