3671 Keratinocytes and Melanocytes Have Distinct and Shared Responses to Metal-Based Nanocatalysts and UVB

E. R. Kisin[1], S. Guppi[1], I. H. Shrivastava[1], V. E. Kagan[2], and A. A. Shvedova[1,3]. *[1]NIOSH, Morgantown, WV; [2]University of Pittsburgh, Pittsburgh, PA; and [3]West Virginia University, Morgantown, WV.* Sponsor: T. Stueckle

Metal-based nanocatalysts (NCT) have an irreplaceable role in energy conversion, chemical production, and automotive exhaust purification. Common metal NCT include transition metals (Me) and alloys, which provide active sites for catalytic reactions. However, the same properties that make these NCT very attractive can pose potential health risks. In addition, interactions of nanoparticles (NPs) with constituent skin cell types, in particular after the cells have been subjected to environmental stress like UVB exposure (considered as the main cause of skin cancer) are essential. In this study, we evaluated the ability of four different NCT ($NiFe_2O_4$, $CoFe_2O_4$, Ni and Co_3O_4) to initiate oxidative stress, induce redox-sensitive transcription factors and trigger inflammatory response in primary human epidermal keratinocytes (HEK) and melanocytes (HEM). Me/MeO-exposed cells, both HEK and HEM, revealed a dose- and time-dependent reduction in viability, cell damage, activation of NF-κB, elevated ROS generation, release of inflammatory mediators, increase in oxidative stress and DNA damage markers. Me/MeO adverse responses were significantly amplified by pre-exposure of both cell types to UVB ($4KJ/m^2$). Hierarchical clustering analysis of inflammatory responses suggested cell-type specific effects as well as treatment related differences. Interestingly, only HEM-exposure to Me/MeO resulted in the amplification of placenta growth factor's (VEGF) response (with and without pre-exposure to UVB). Notably, VEGF plays a crucial role in melanoma invasiveness, vasculogenic mimicry and tumor-associated angiogenesis. HEK pre-exposure to UVB induced expression of IL-17, a pro-inflammatory cytokine, associated with hyperproliferation and abnormal differentiation of keratinocytes. Our results indicate that topically applied metal NP may exert differential cellular effects on UVB exposed skin which are important in the context of neoplastic transformations.

3672 Size and Coating-Dependent Toxicity and Pro-inflammatory Effects of Silver Nanoparticles in Human Monocytes and Macrophages

E. Fernandes, and F. Carvalho. *Universidade do Porto, Porto, Portugal.*

There is a growing concern about the increased human exposure to silver nanoparticles (AgNP) and their potential harmful effects. When AgNP reach the bloodstream, interact with "guard cells" of the immune system, contributing to the onset and development of an inflammatory response. Monocytes and macrophages play a pivotal role in our defense system, being both involved in the regulation of the initiation, development, and resolution of inflammation. The objective of this work was to assess the pro-inflammatory effects induced by AgNP of different sizes (5, 10 and 50 nm), and capped with two coating agents [polyvinylpyrrolidone (PVP) and citrate], in human monocytes isolated from human blood and human macrophages derived from a monocytic cell line (THP-1). Several steps of the inflammatory cascade were assessed including the interference with cells viability, production of reactive pro-oxidant species, depolarization of mitochondrial membrane potential, and cytokines and chemokines release (IL-8, IL-1β, IL-6, IL-10, TNF and IL-12p70). It was observed that the studied AgNP exert deleterious effects in both cell types. On human monocytes, it was observed an induction of late apoptosis, more exuberant for smaller AgNP with the two tested coatings. On human macrophages, it was observed the occurrence of apoptosis, late apoptosis and necrosis. While apoptosis and late apoptosis were induced by the smaller sizes of PVP and citrate-coated AgNP (5 and 10 nm), necrosis was only induced by PVP and citrate-coated 50 nm AgNP. No effect was observed on pro-oxidant species production, irrespective of size and coating. Human monocytes exhibited an exuberant depolarization of mitochondrial membrane potential, which was dependent on the concentration, but not on size and/or coating, while no effects were observed in human macrophages. Interestingly, a significant production of anti-inflammatory IL-10 and pro-inflammatory cytokines, namely TNF, IL-8 and also IL-6 was observed in both cellular models. In general, these results showed that human monocytes seem to be more sensitive to AgNP exposure than human macrophages. This work prompts us to further explore the adverse outcomes and mechanistic pathways leading to AgNP-induced pro-inflammatory effects. *The present work was supported by UID/QUI/50006/2020 and COMPETE (PTDC/NAN-MAT/29248/2017-POCI010145FEDER029248). AS thanks FCT and ESF through POCH for her PhD grant reference SFRH/BD/150656/2020. ATR thank to FCT for the funding through the project PTDC/MED-QUI/29243/2017. MF acknowledges her contract under the Scientific Employment Stimulus - Individual Call 2020.04126.CEECIND/CP1596/CT0006.*

3673 Neoplastic Transformation and Changes in Transcriptome Induced by Riebeckite/Tremolite Asbestiform and Non-asbestiform Elongate Mineral Particles in Human Mesothelial Cells

A. A. Shvedova[1,2], S. Guppi[1], I. H. Shrivastava[1], T. O. Khaliullin[3], N. Yanamala[4], E. R. Kisin[1], and V. E. Kagan[5]. *[1]NIOSH, Morgantown, WV; [2]West Virginia University, Morgantown, WV; [3]Ohio State University, Columbus, OH; [4]Rutgers Robert Wood Johnson Medical School, Newark, NJ; and [5]University of Pittsburgh, Pittsburgh, PA.* Sponsor: T. Stueckle

Inhalation of respirable amphibole asbestiform elongate mineral particles (EMPs) can cause asbestosis, lung cancer as well as pleural and peritoneal mesothelioma. Amphiboles can also occur in a non-asbestiform habit that can be mechanically broken into so-called cleavage fragments (CF) which can meet the mineralogical/regulatory criteria for fibers. While the health effects of riebeckite (RF) and tremolite (TF) asbestos are well documented, there is uncertainty regarding the toxicity of their CF. In this work, we evaluated several neoplastic-like transformation hallmarks and changes in the gene expression profiles in immortalized human mesothelial (MET-5A) cells after continuous long-term exposure to sub-toxic ($2.5\ \mu g/cm^2$) concentrations of RF, TF and corresponding CF. TF- and RF-exposed MET-5A cells acquired a neoplastic-like transformed phenotype characterized by significant morphological changes accompanied by an increase in anchorage-independent growth (AIG), invasion/migration and proliferation. No AIG was observed in cells treated with riebeckite and tremolite CF, although an increase in migration and invasion parameters were detected. Hierarchical cluster analysis and ingenuity pathway analysis revealed altered gene expression profiles common for asbestos fibers and corresponding cleavage fragments. RF and riebeckite CF had enriched pathways pertaining to cellular development, cellular growth, proliferation, as well as cancer and organismal injury disease categories. RF exposure alone induced pronounced upregulation of genes governing cell-to-cell signaling and interactions related to cell death and survival. Genes regulated by TF and its CF were also associated with cell-to-cell signaling and interactions, cell development, growth and proliferation, whereas only TF caused enrichment of immune cell trafficking and cancer-related pathways. In conclusion, at equal mass-based doses, asbestiform amphiboles had greater potential for the neoplastic transformation of mesothelial cells than corresponding CF. Discrete mineral-specific responses were also induced in MET-5A cells by non-asbestiform EMPs.

3674 Biotransformation Modulates the Penetration of Metallic Nanomaterials across the Blood-Brain Barrier

Z. Guo, P. Zhang, and I. Lynch. *University of Birmingham, Birmingham, United Kingdom.*

Although brain is protected by a tight physiological guardian named blood brain barrier (BBB), deposition of engineered nanomaterials (ENMs) in brain and consequent neurotoxicity has been reported. To date, it is still unclear whether and how ENMs enter the brain by crossing the BBB. Understanding the potential of ENMs to cross the BBB as a function of their physicochemical properties and subsequent behavior, fate, and adverse effect beyond that point is vital for evaluating the neurological effects arising from their unintentional entry into the brain, which is yet to be fully explored. This is not only due to the complex nature of the brain but also the existing analytical limitations for characterization and quantification of NMs in the complex brain environment. Herein, we conducted an interdisciplinary study by using a novel analytical workflow and *in vitro* BBB model, as a complex biological barrier, to determine and quantify the biotransformation of metallic NMs as a function of their physicochemical properties and correlate the influence of the biotransformation to the BBB-penetration ability and transport pathways. We found metallic ENMs transform in the BBB as affected by their shape, size and intrinsic solubility, which in turn modulates their transport form, efficiency and pathways through the BBB, and consequently their neurotoxicity. Very little was transcytosed to the basolateral (brain) side of the BBB model, with significant amounts being recycled back to the apical (bloodstream) side and limited retention in the BBB cells. Paracellular transport was only observed at the higher concentration tested and was associated with membrane damage and NM dissolution. The generated data about biotransformation modulated uptake and transport of NMs through BBB open a new horizon for medical application of NMs, e.g. targetable drug delivery systems for brain diseases and also for biological fate assessment of NMs in brain to support their risk assessment.

3675 Exposure to MgO Food Additives Affects Human Commensal Bacteria

A. Garcia-Rodriguez[1], A. Stillwell[1], B. Tochilovsky[1], R. Limage[1], J. V. Tanzman[1], N. Kolba[2], E. Tako[2], C. Marques[1], and G. Mahler[1]. *[1]University of Binghamton, Binghamton, NY; and [2]Cornell University, Ithaca, NY.*

The gastrointestinal (GI) microbiota is linked to intestinal homeostasis and is crucial for overall host health. When the microbiota is impaired or altered, common inflammatory and metabolic disorders are more likely to develop. Exogenous factors such as diet have been reported to highly impact microbial dysbiosis.

SOT 61ST ANNUAL MEETING & TOXEXPO • SAN DIEGO, CA MARCH 27–31, 2022

The Toxicologist

Supplement to *Toxicological Sciences*

SOT | Society of Toxicology

Toxicological Sciences
The Official Journal of the Society of Toxicology
www.academic.oup.com/toxsci

OXFORD UNIVERSITY PRESS

ISSN 1096-6080 Volume 186, Issue S1 March 2022

Publication Date: March 23, 2022