of 12 types (40, 60, 80 and 100 nm coated with citrate, polyvinylpyrrolidone (PVP), and branched polyethyleneimine (BPEI)) of AgNPs on organelle morphology in human retinal pigmented epithelial (ARPE-19) cells. AgNPs (0.1-30 µg/ml) were applied to the cells seeded in a 384-well format, with silver nitrate acting as silver ionization control. The Distorted Grid (DG) model was used to estimate the cellular delivered dose. After 24 hrs of treatment, cells were live-labeled with MitoTracker (mitochondria), fixed, permeabilized and labeled with Hoechst-33342 (nuclei), SYTO14 (nucleoli) and fluorescent conjugates of concanavalin A (ER), phalloidin (actin cytoskeleton), and wheat germ agglutinin (Golgi/plasma membrane). A multiplexed cell viability and apoptosis assay was run in parallel. Cells were imaged using an Opera Phenix High Content Screening System and profiled using Harmony High Content Analysis software. Approximately 1200 morphological features were measured per cell and summarized to the well level for analysis. The DG model predicted that the fraction of AgNP deposited on cells increased with particle size and differed based on coating (BPEI > citrate > PVP). Phenotypic profiling showed that all AgNP types affected the organelle morphology in a concentration-dependent manner, with over 600 features having benchmark doses below the threshold for cytotoxicity. The pattern of changes in cell morphology differed across coating agents and silver nitrate. Citrate coated AgNPs showed the most pronounced effects below the cytotoxic threshold. 60 nm PVP had fewer effects on cell morphology than other coatings, yet apoptosis was observed at an estimated delivered dose as low as 1.45 µg/ml. This screening method may inform subsequent assay selection by highlighting the intracellular regions affected by AgNPs of interest. *This abstract does not necessarily reflect US EPA policy.*

2200 Amorphous Silica Coating Protects against Iron Oxide Nanoparticle-Induced Cell Transformation and Genotoxicity

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Iron oxide nanoparticles (IONP) have a wide range of uses in biotechnology, medicine, and transportation. However, very little is known about their potential adverse health effects following human exposure. Some evidence suggests that dissolution of IONP following endocytosis into cells may disrupt iron homeostasis, resulting in genotoxicity and neoplastic-like cellular transformation. Surface modification of IONP, such as an amorphous silica coating, may impact subsequent adverse outcomes by reducing particle dissolution. The main objective of this study was to assess IONP low dose, long term exposure effects, including carcinogenic potential, as well as the utility of an amorphous silica coating in reducing or preventing these outcomes. Human bronchial epithelial cells (Beas2B) were continuously exposed to nFe_2O_3 or nano-SiO_2 coated nFe_2O_3 (SiO_2-nFe_2O_3) for up to 6.5 months at an occupationally relevant low dose (0.6 µg/cm^2 or 2.88 µg/mL) and evaluated over time for indications of neoplastic-like transformation and its underlying mechanism. Transformation was compared to that induced by gas metal arc mild steel welding fumes (GMA-WF), which were recently re-classified as a Group 1 total human carcinogen, and are composed of roughly 80% iron/iron oxide. Our results showed that beginning at four months, nFe_2O_3-exposed Beas2B underwent neoplastic-like transformation, as indicated by increased cell proliferation and attachment-independent colony formation. These outcomes correlated with nFe_2O_3 dissolution, increased intracellular iron, and genotoxicity, as well as significant changes in pathways related to DNA damage repair and autophagic processes. nFe_2O_3-induced transformation also closely matched that GMA-WF induced transformation SiO_2-nFe_2O_3 treatment, however, did not induce any changes in the above parameters. Overall, our results indicated potential carcinogenic risk of nFe_2O_3 associated with particle dissolution, iron homeostasis disruption, and changes in autophagy and DNA damage repair pathways, which were reduced with an amorphous silica surface coating. This study shows the potential utility of a "safe by design" hazard reduction strategy, to alter particle physicochemical properties based on mode of toxicity to reduce risk.

2201 Potential Role of Thioredoxin-Interacting Protein in Silver Nanoparticle-Mediated Mast Cell Degranulation

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Silver nanoparticles (AgNPs) are being incorporated into a variety of consumer and medical products primarily due to their antimicrobial properties. This will lead to a significant rise in exposure to the general population; however, our understanding of the potential adverse health effects of AgNPs is still minimal. We have previously demonstrated that AgNPs induce a non-IgE mediated degranulation of mast cells. Importantly, we have also shown a strong genetic contribution to AgNP driven mast cell degranulation. RNA sequencing performed on bone marrow-derived mast cells (BMMCs) from high-responding (C57BL/6) and low-responding (LP/J) strains of mice demonstrated a significant increase in thioredoxin-interacting protein (*txnip*) in the low-responding strain after exposure to 20nm AgNP. We therefore explored the role of TXNIP in mast cells to determine its potential regulatory role in AgNP-mediated degranulation. At one hour following exposure to AgNP, *txnip* mRNA levels were increased in LP/J but not C57BL/6 which confirms RNA sequencing results. Protein levels of TXNIP were similarly increased in LP/J BMMCs at six hours post-exposure while protein levels were significantly decreased in C57BL/6 BMMCs. siRNA knockdown of TXNIP resulted in a trend towards increased mast cell degranulation. Using a Seahorse XF analyzer, we found that BMMCs from low- and high-responding strains possess varying glycolytic capacities in response to AgNP exposure possibly implicating TXNIP as a regulatory protein for cellular metabolism during mast cell degranulation. Our data suggests that TXNIP plays a role in non-IgE mediated mast cell degranulation initiated by exposure to 20nm AgNP and may possibly modulate cellular metabolism.

2202 *In Vitro* Dermal Toxicity of Redox-Active Metal Nanocatalysts

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Nanocatalysts (NCT) represent the convergence of catalysts, a mature technology with a new one, nanotechnology. NCT is a rapidly growing field that involves the use of nanomaterials as catalysts for a variety of catalytic applications. Since metal nanoparticles (MeNP) have a large surface-to-volume ratio compared to bulk materials, they are attractive candidates for use as catalysts. A number of redox-active MeNP and their oxides (MeO) including nickel (Ni) and cobalt (Co) are widely used. The physical nature and reactive surface properties of some of these may affect their ability to induce dermal toxicity thus causing adverse skin reactions. We hypothesize that toxicity of Me/MeO NP occurs via their ability to initiate oxidative stress, thereby inducing redox-sensitive transcription factors and triggering inflammation. Moreover, due to the skin's susceptibility to UV radiation, it is important to evaluate whether Me/MeO NP enhance the adverse effects of UVB. To test these hypotheses, the effects of Ni, Co, NiO, Co_3O_4 and CoO alone (0-26 µg/cm^2) and co-exposed with UVB (4KJ/m^2) were studied *in vitro* and *in situ* using murine epidermal cells (JB6 P+/+) and an engineered human skin construct (EpiDerm FT). Cell exposure to Me/MeO NP resulted in a dose- and time-dependent loss of cell viability, cell damage, oxidative stress and activation of AP-1/NF-κB. Co-exposure of Me/MeO NP with UVB ensued in amplification of the observed effects. Exposure of EpiDerm FT to Me/MeO NP caused tissue damage, oxidative stress and accumulation of inflammatory mediators. Hierarchical cluster analysis resulted in two major clusters separating cytokines production related to inflammatory cell recruitment (more intense) and T_H2-type/regulatory immune responses (dimmed). UVB exposure alone induced significant tissue damage and secretion of cytokines/chemokines. Ni compounds drastically enhanced the post-UV treatment LDH release and secretion across the whole cytokine spectrum, while Co oxides prompted much weaker reaction. Interestingly, inflammatory cytokine/chemokine levels upon exposure to Me/MeO NPs, with or without UVB pre-treatment, followed similar trends compared to cell/tissue damage i.e., NiO>Ni>Co_3O_4>CoO and correlated with their size. Altogether, these results clearly indicate that some of the Me/MeO NP could induce cytotoxicity, oxidative stress and inflammation, and may potentially enhance response caused by UVB pre-treatment. *Disclaimer: The findings and conclusions of this report are those of the authors and do not necessarily reflect those of National Institute for Occupational Safety and Health.*

2203 Comparative *In Vitro* Study of Adverse Pro-neoplastic Potential of Tremolite Asbestos and Its Cleavage Fragments in Human Epithelial (BEAS-2B) and Mesothelial (MET-5A) Cells

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The fibrous form of tremolite is one of the six recognized types of asbestos. It is known that inhalation of respirable tremolite fibers (TF) can cause asbestosis, lung cancer and both pleural and peritoneal mesothelioma. Tremolite also occurs in a non-fibrous habitat that can be mechanically broken into cleavage fragments (CF) which can meet the criteria for fibers. Despite the considerable amount of work showing that CF are less potent in their damaging effects than asbestos fibers, little data exists on the adverse effects of well-characterized tremolite CF. The present study was designed to reveal the potential pro-carcinogenic manifestations of respirable TF and correspond-

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