difference in agglomeration between the two nanoparticles types. In addition, the PS-Ag is more biocompatible than the HC-Ag however exposure to the alveolar, interstitial, and lysosomal fluid had a stimulatory effect on the cells with an increase in viability observed. In contrast, over time exposure to the gastric fluid made the nanoparticles more toxic in a dose-dependent manner. This study demonstrated when addressing nanotoxicity, the effect of the aqueous biological environment not only effects the dynamics of aggregation and agglomeration but also modulates the presentation and exposure properties of nanomaterials on cellular and tissue systems.

PL 1974 DOSE AND RESPONSE METRICS IN ASSESSING *IN VITRO* AND *IN VIVO* NANOPARTICLE TOXICITY.

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The increasing number and variety of new nanoparticles (NPs) calls for the development of low-cost in vitro screening assays for ranking NP toxicity. Unlike chemicals which are usually measured by mass dose, the great variety of physicochemical properties of NPs (size, shape, crystallinity, etc) means that other parameters need to be scrutinized in order to examine how useful these in vitro assays are in predicting the in vivo toxicities. We explored several dose and response metrics with the objective to predict in vivo toxicity from in vitro data. Critical to this effort are the choices of biological endpoints and cellular systems that are relevant targets in vivo. We assessed toxicity of several TiO2 NPs of sizes 3-100 nm. A rat lung Type I epithelial cell line (R3/1) was used for in vitro study; in vivo study involved intratracheal instillation of NPs to rats. In vitro endpoints were lactate dehydrogenase (LDH) release, protein carbonylation, and heme oxygenase-1 (HO-1). Number of PMNs in bronchoalveolar lavage fluid (BALF) was used as endpoint in vivo. Several metrics were used to rank the toxicity of these NPs, including ED50 and the steepest slopes in the dose-response curve calculated using two different methods. We then compared the correlations between the in vitro and in vivo ranking results. Carbonylation and HO-1 assays were good markers of oxidative stress related toxicity. The steepest slope in the dose-response curve was the best response metric for ranking NP toxicity when both dose and response were expressed in unit surface area. It was concluded that toxicity rankings by certain in vitro assays were consistent with the in vivo toxicity rankings when proper dose and response metrics were utilized. [US DOD MURI grant FA9550-04-1-430; UofR NIEHS Toxicology Training Grant T32 ES07026].

PL 1975 MECHANISMS OF INHALED MULTIWALLED CARBON NANOTUBE-INDUCED IMMUNOSUPPRESSION.

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Inhalation of multiwalled carbon nanotubes (MWCNT) showed decreased systemic immune function with is modulated by the COX-2 pathway. These studies report on elucidation of this pathway. Male mice were exposed to atmospheres containing 0.3 or 1 mg/m3 MWCNT for 6 hours/day for 14 consecutive days in whole body inhalation exposure chambers. Approximately 18 hours after exposure mice were assessed for systemic immune function in the spleen. Systemic immune function was compromised after 1mg/m3 MWCNT exposure. Splenocytes from exposed animals were less able to produce antibody in response to antigenic stimuli and exhibited decreased T cell proliferation when co-cultured with a mitogen (Concanavalin A). Furthermore, splenocytes from exposed animals exhibited increased gene expression of prostaglandin synthase enzymes. Prostaglandin synthase enzymes catalyze the metabolism of arachidonic acid to prostaglandins; known T cell suppressors. Therefore an additional inhalation exposure was conducted with mice that received a Prostaglandin Synthase 2 (PTGS2 or COX-2) antagonist (ibuprofen) in their drinking water. Ibuprofen treated animals exhibited significant rescue from MWCNT-induced immunosuppression, suggesting involvement of prostaglandins in immune function alterations. Subsequent experiments showed COX-2 (PTGS2) knockout mice were resistant to inhaled MWCNT-induced immune alterations. Finally, co-culturing naïve splenocytes with protein collected from lung washes in MWCNT exposed mice showed similar responses as what was observed in vivo. This finding suggests the effects arise from a signal in the lung, not from translocation of the MWCNT to the spleen. This work was supported by NIEHS (P30 ES-012072) and EPA (RD-83252701).

PL 1976 NADPH OXIDASE REGULATES NEUTROPHILS AND FIBROSIS IN C57BL/6 MICE EXPOSED TO CARBON NANOTUBES.

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Single-walled carbon nanotubes (SWCNT) have been introduced into a large number of new technologies and consumer products. The combination of their exceptional features with very broad applications raised concerns regarding their potential health effects. The prime target for SWCNT toxicity is believed to be the lung where exposure may occur through inhalation, particularly in occupational settings. Our previous work has demonstrated that SWCNT cause robust inflammatory responses in rodents with very early termination of the acute phase and rapid onset of chronic fibrosis. Timely elimination of polymorphonuclear neutrophils (PMNs) through apoptosis and their subsequent clearance by macrophages is a necessary stage in the resolution of pulmonary inflammation whereby NADPH oxidase contributes to control of apoptotic cell death and clearance of PMNs. Thus, we hypothesized that NADPH oxidase may be an important regulator of the transition from the acute inflammation to the chronic fibrotic stage in response to SWCNT. To experimentally address the hypothesis, we employed NADPH oxidase-deficient mice which lack the $gp91^{phox}$ sub-unit of the enzymatic complex. We found that NADPH oxidase null mice responded to SWCNT exposure with a marked accumulation of PMNs and elevated levels of apoptotic cells in the lungs, production of pro-inflammatory cytokines, decreased production of the anti-inflammatory and pro-fibrotic cytokine, TGF-β, and significantly lower levels of collagen deposition, as compared to C57BL/6 control mice. These results demonstrate a role for NADPH oxidase-derived reactive oxygen species in determining course of pulmonary response to SWCNT. Acknowledgements: supported by NIOSH OH008282, NIH HL70755, NORA 927000Y, and the 7th Program of the EC (EC-FP-7-NANOMMUNE-214281).

PL 1977 MODELING MOLECULAR INTERACTIONS BETWEEN MARCO AND NANOSILICATES.

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Airborne exposure to environmental particulates is associated with inflammation and adverse health effects, in particular increased pulmonary and cardiovascular morbidity and mortality. Scavenger receptors, expressed on the cell surface of macrophages, have been implicated as responsible for recognition and internalization of micron-sized environmental particles. However, the molecular mechanism of engineered nanoparticles recognition and uptake has not been addressed. Recently, the three-dimensional structure of the cysteine-rich domain (SRCR) of the macrophage receptor with collagenous structure (MARCO) has been determined by X-ray crystallography. The SRCR domain of MARCO was shown to be involved in cell interactions with LTA, LPS, intact bacteria, and some metal oxide particulates. In this work, the binding of silicates to MARCO is characterized through computer simulations involving its SRCR domain and a number of silicates. Molecular docking of a variety of nanowire silicates were used as screening probes to identify the putative nanosilicate binding sites on MARCO surface. Preferred binding sites involve the arginine clusters and the dimer interface. While the latter only is accessible to sub-nanoparticles, larger silicates can bind to the same region identified to bind LPS. This information has been subsequently used to model the interaction of the receptor to a fully hydroxylated (010) silica surface. Molecular dynamics simulations reveal that the binding is an energetically favorable process and involves the arginine cluster at the -sheet outer surface. Although the secondary structure of MARCO is maintained, significant rearrangement at the dimer interface was observed upon substrate adhesion slightly altering MARCO electrostatic surface potential signature. These results highlight, for the first time, potential differences between MARCO interactions between micron and nanoscale particles, and suggest potential receptor structural rearrangements that may facilitate nanoparticle induced signaling in macrophages. Supported by R01 ES016212 and the Environmental Biomarkers Initiative.

PL 1978 DIFFERENTIAL GENOTOXIC EFFECTS OF TRANSITION METAL NANOPARTICLES: THE ROLE OF OXIDATIVE STRESS.

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Nanotechnology is considered to be one the world's most promising new technologies. It directly improves our lives in areas as diverse as engineering, information technology, and diagnostics. Our current knowledge of the health effects of metal

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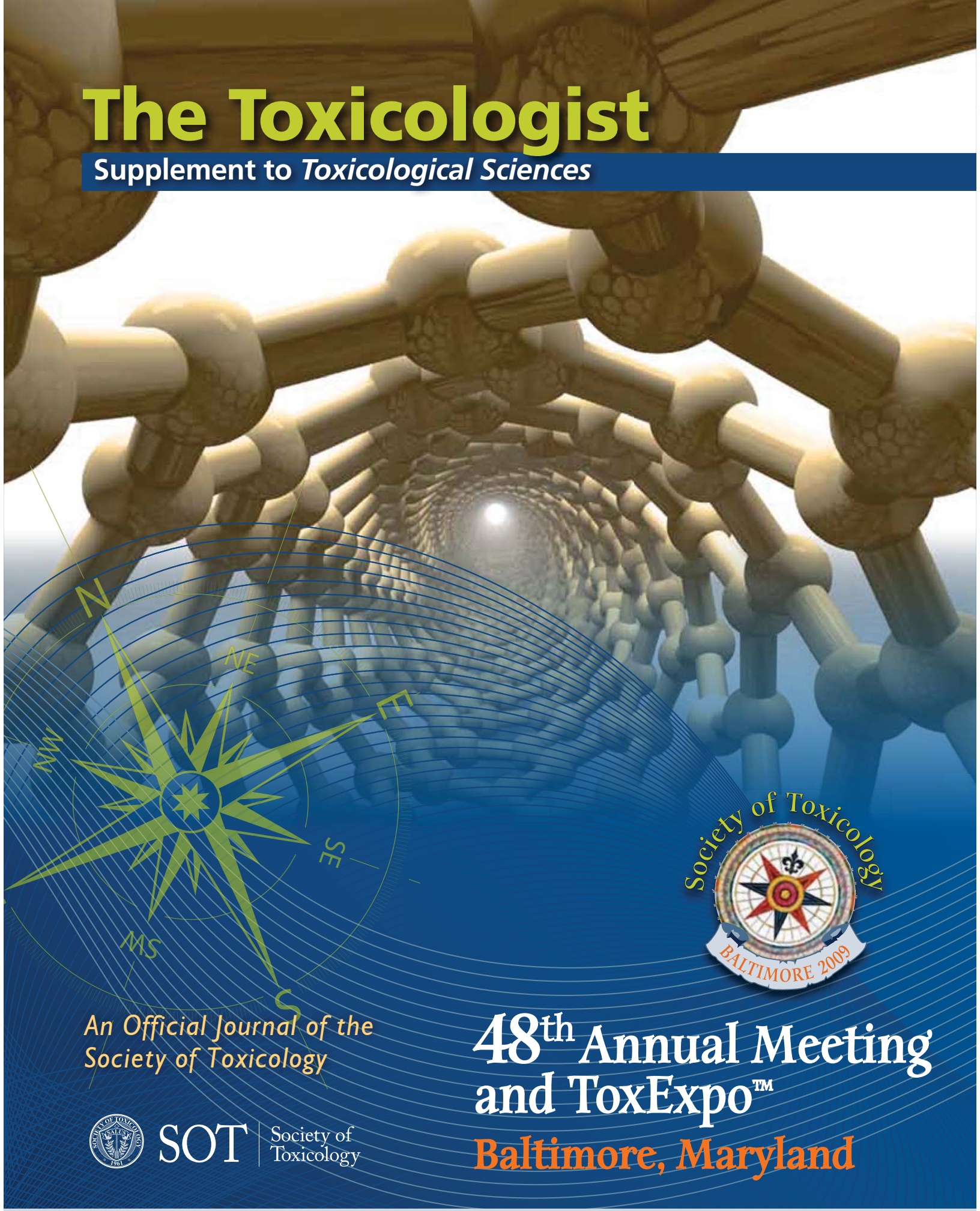

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