been reported, the mechanism of their toxicity is uncertain. In this study, we seek to understand the toxic mechanisms of nanoparticles using an *in vitro* model and relevant biological endpoints to provide a foundation to further investigate the toxicity of nanotubes *in vivo*. Representative fullerene and carbon nanoparticle samples were obtained and characterized, including impurity profiles. BEAS-2B cells were exposed to different doses of nanoparticles ranging from 0.1ug/ml to 1000 ug/ml suspended in DMEM medium by sonication. Cell viability measurements using neutral red uptake and trypan blue exclusion, and nuclear factor kB (NF-kB) activation measurement by luminometer were conducted immediately after 24 hrs of exposure. The in-vitro study shows that the combustion fullerene soot (98% fullerene) produced greater NF-kB activation than Arc fullerene soot (7% fullerene), while both of these particles produced greater response than fine or ultrafine TiO2, and less than that produced by crystalline SiO2. Based on the dose-response obtained by in-vitro study, each kind of particles were intratracheally instilled to CAF1/J mice at 100 and 250 ug/mice suspended in 50 ul 1% DMSO in normal saline. Lung lavage parameters were measured 24 hrs post exposure. In both arc and combustion fullerene exposed mice at both doses, total cell counts and protein levels were elevated comparing to saline-exposed control mice, although no differences were observed between arc and combustion fullerene exposed mice for these parameters. These data suggest that exposure to fullerene soot may lead to pulmonary toxicity and that the response appeared to depend on the types of nanomaterial used and their impurities.

1038 EXPOSURE TO ULTRAFINE ELEMENTAL CARBON PARTICLES (UCP) SIGNIFICANTLY INCREASE THROMBOGENESIS.

V. M. Silva, N. Corson, A. Elder, R. Gelein and G. Oberdorster. *Environmental Medicine, University of Rochester, Rochester, NY.*

Recent studies indicate that exposure to ultrafine particles (UFP) can increase thrombogenesis. We have established a non-invasive model of *in vivo* thrombo-genesis in ear veins of rats. We have shown with this model that intravenous (IV) and intratracheal (IT) administration of ultrafine aminated-polystyrene particles significantly induced thrombus formation. The objective of this study was to determine if environmentally-relevant UFP such as ultrafine elemental carbon particles (UCP), affect coagulation when administered to rats through IV, IT and inhalation routes. For this purpose, UCP were generated as an aerosol with count median diameters (CMDs) of 25-35 nm (GSD=1.7) using electric spark discharge of ultrapure graphite electrodes in argon. Particles were collected on filters, and then suspended in saline (250 µl) at various concentrations (4, 20, 100 and 500 g/kg) for IV or IT administration into rats. In inhalation studies animals were exposed to UCP for 30 min to 3 hrs in whole-body inhalation chambers at concentrations of 200 and 70 µg/m^3. Our data show that IV doses as low as 4 µg UCP/kg (~1g/rat) significantly shorten the time of thrombus formation in rat ear veins. Although the effect of the different IV doses is not significantly different from each other, there seems to be a trend indicating that the lower the dose of UFP in the system the better the response. Similar results were obtained when particles were IT instilled into rats. In this case, even a lower dose (0.8 µg/kg or 0.2 µg/rat) of UCP enhanced thrombus formation. Inhalation of 25-35nm carbon particles significantly induced coagulation regardless of the dose. Interestingly, this response did not change with time of exposure. These results are consistent with the hypothesis that UCP deposited in the lung translocate to the blood circulation and can activate platelets directly. Furthermore, these data demonstrate that the non-invasive ear vein model is useful to study UFP-induced thrombogenic effects after inhalation exposure.

1039 NANOPARTICLE DEPOSITION EFFICIENCY IN HUMAN NASAL AIRWAY REPLICAS.

B. A. Wong[1], J. T. Kelly[2], J. S. Kimbell[1] and B. Asgharian[1]. *[1]CIIT CHR, Research Triangle Park, NC and [2]University of California, Davis, CA.*

Particles in the nanometer size range (1-100 nm) may be released to the environment by various means, including escape during manufacturing processes, production during combustion processes, and formation by condensation from gaseous precursors. Toxicologists have long been concerned with, and have studied the respiratory toxicology of particles in the micrometer and submicrometer size range. However, less is known about potential toxicity associated with nanoparticles. The overall deposition efficiency of nanoparticles in the nasal airways provides information about the relative dose between the upper and lower respiratory tract. We studied nanometer particle deposition efficiency in plastic replicas of nasal airways and compared our results with other models. Two replicas were manufactured using stereolithography techniques with morphological data from an MRI scan of a human nose. Monodisperse particles of nanometer and ultrafine sizes from 5 to 150 nm were generated into a constant air flow of 10 and 20 L/min. Deposition efficiency was determined by measuring the particle concentration at the entrance and outlet of the nasal replica. Deposition efficiency was less than 10% for particles > 30 nm and increased for particles < 30 nm. The increase was attributed to increased particle diffusivity. Our deposition measurements were comparable with deposition values reported for other models. These results suggest that differences in nasal airway morphometry do not significantly affect the overall deposition efficiency of nanometer-sized particles.

1040 ELECTRON MICROSCOPIC STUDY ON THE TRANSLOCATION OF ULTRAFINE CARBON BLACK PARTICLES AT THE AIRWAY-CAPILLARY BARRIER IN LUNG.

A. Shimada, N. Kawamura, T. Kaewamatawong, M. Okajima, M. Sawada and T. Morita. *Veterinary Pathology, Tottori University, Tottori-shi, Japan.*

Ultrafine particles (UFP) may induce adverse respiratory and cardiovascular effects. Inhaled UFP have been shown to translocate to systemic circulation. Precise mechanisms of the anatomical translocation (crossing the airway-capillary barrier) of inhaled UFP are not fully understood. We examined anatomical location of the intratracheally instilled UFP at light and electron microscopic levels. Ultrafine carbon black (UFCB) particles, printex 90 (Degussa, Frankfurt, Germany), 14nm in diamiter, were instilled to the trachea of 10-week-old ICR female mouse at a concentration of 1mg/0.05ml/body. Lung, regional pulmonary lymph nodes, liver and spleen were removed at 0, 5, 10, 30 min, 1, 2, 6, 12 and 24 hrs after instillation (n = 3 at each time point). Paraffin sections cut at 2um were processed for Factor VIII-immunohistochemistry to define blood vessels. Formalin-fixed lung samples were processed for electron microscopy. Aggregates of UFCB were observed in the capillaries of the alveolar walls soon after instillation. UFCB particles, which confined to the cytoplasm of the mononuclear cells with morphological appearance of dendritic cells, appeared in the regional pulmonary lymph nodes at 24 hrs after instillation. No UFCB particles were observed in the liver and spleen. Electron microscopy demonstrated aggregates of UFCB between the edges (epithelial pores) of the elongated thin cytoplasm of type I alveolar epithelial cells. Endothelial cells of the alveolar capillaries appeared to be highly activated with extensive folding, a large number of pinocytic vesicles and ribosomes. The thickness of the basement membrane (BM) varied from point to point. Occasional UFCB were observed in the matrix of the BM, pinocytic vesicles and in the spaces between the folded cytoplasm (fenestration) of the endothelial cells. These results suggest that pro-inlfammatory chemokines responsible for the morphological alterations observed may be involved in the passage of the instilled UFCB through the airway-capillary barrier.

1041 PULMONARY TOXICITY OF CARBON NANOTUBES.

E. Kisin[1], A. R. Murray[3], V. Johnson[2], O. Gorelik[4, 5], S. Arepalli[4, 5], V. Z. Gandelsman[4, 5], A. F. Hubbs[1], R. R. Mercer[1, 3], P. Baron[6], V. E. Kagan[7], V. Castranova[1, 3, 7] and A. A. Shvedova[1, 3]. *[1]PPRB, HELD/NIOSH, Morgantown, WV, [2]TMBB, HELD/NIOSH, Morgantown, WV, [3]Physiology & Pharmacology, WVU, Morgantown, WV, [4]Lockheed Martin Corporation, Engineering Directorate, Material & Processes Branch, NASA-JSC, Houston, TX, [5]Nanotube Team, GBTech, Inc., NASA-JSC, Houston, TX, [6]MRSA/DART, NIOSH, Cincinnati, OH and [7]University of Pittsburgh, Pittsburgh, PA.*

Carbon nanotubes (CNT) are new members of carbon allotropes similar to fullerenes and graphite. Because of their unique electrical, mechanical and thermal properties, carbon nanotubes are being evaluated for novel applications in the electronics, aerospace and computer industries. Previously, we have observed that exposure of human bronchial epithelial cells to CNT induced iron-dependent oxidative stress, depletion of antioxidants, morphological changes, cytotoxicity, and apoptosis. In the current study, we investigated pulmonary toxicity of CNT in C57BL/6 mice after pharyngeal aspiration. End points were examined on days 1, 3, 7, 14, 28, and 60 post-exposure. We found that CNT caused dose-dependent formation of granulomatous bronchointerstitial pneumonia, fibrosis, and altered pulmonary function. Administration of CNT to C57BL/6 mice also resulted in a dose-dependent augmentation of inflammation biomarkers quantified by cell counts, total protein, lactate dehydrogenase (LDH) andγ-glutamyltranspeptidase (GGT) activities in bronchoalveolar lavage (BAL) fluid samples. Markers of pulmonary cytotoxicity were associated with the development of inflammation, collagen accumulation, and pulmonary fibrosis. TGF-β was maximally increased in BAL fluid of mice 7 days after CNT exposure and correlated with morphometric evidence of collagen formation as well as pulmonary function changes. Mice exposed to an equal mass of ultrafine carbon black or fine crystalline silica exhibited less PMN recruitment and cytotoxicity than mice receiving CNT. Our data suggest that exposure to CNT leads to pulmonary toxicity involving inflammation and oxidative stress, which culminates in the development of multifocal granulomatous pneumonia and fibrosis.

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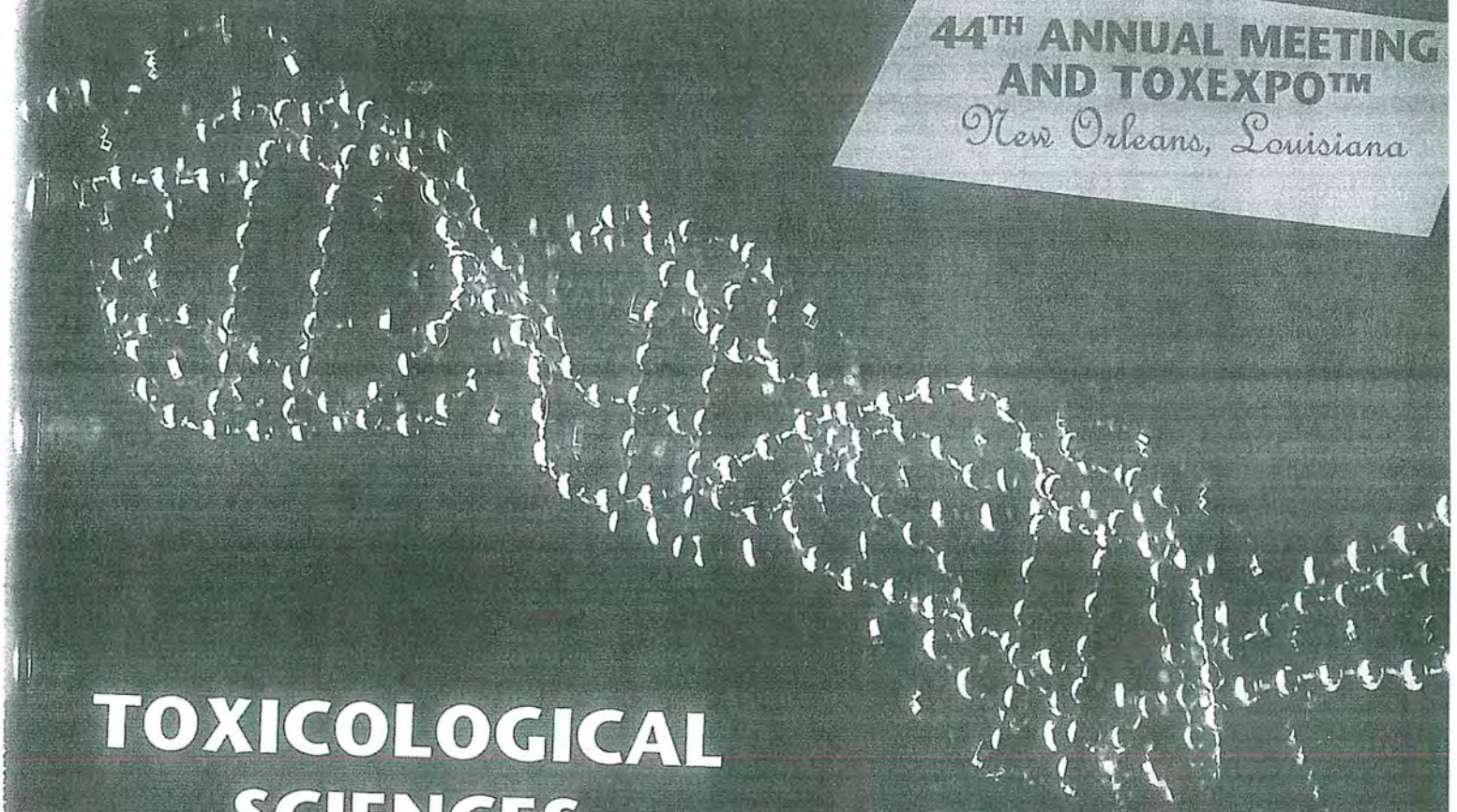

TOXICOLOGICAL SCIENCES

The Official Journal of the Society of Toxicology

Supplement

OXFORD UNIVERSITY PRESS

ISSN 1096-6080
Volume 84, Number S-1, March 2005

www.toxsci.oupjournals.org