pathway that aims to regulate inflammatory processes and compensate apoptotic changes. Overall observed immunological responses related to MWCNT exposure are considered the result of the synergistic effect of systemic (mediated by cells of the exposure routes) and local inflammation (blood cells).

2707 Modulating Redox Status of Carbon Nanodots in THP-1 Human Monocytes

Z. Jia[3], N. H. Chiu[3], Q. Hu[2], S. Khan[3], J. Biswas[3], H. Zhu[1], and R. Y. Li[1]. *[1]Campbell University School of Osteopathic Medicine, Buies Creek, NC; [2]University at Buffalo, Buffalo, NY; and [3]University of North Carolina at Greensboro, Greensboro, NC.*

Carbon nanodots, a new class of carbon nanomaterials with sizes below 10 nm, have recently attracted wide attention due to their superiority in water solubility and wide applications in energy, biological labeling, bioimaging and drug delivery. Monocytes are known to be involved in inflammable vascular diseases, and have been suggested to be the targets for carbon nanodots exposure. Using FITC-Annexin V/7-AAD flow cytometry this study for the first time demonstrated that carbon nanodots at concentrations of 0.3-1.2 mg/mL for 6 h elicited no toxic effects to THP-1 human monocytes. The exposure to carbon nanodotsat such concentrations showed no significant differences in the levels of cellular glutathione and glutathione S-transferase in the cells. However, carbon nanodots have shown to scavenge superoxide and peroxynitrite species in a dose-dependent fashion. Also the pretreatment of cells with carbon nanodots afforded a remarkable protection against acrolein-induced cell deaths as determined by FITC-annexin V and 7-AAD flow cytometry. Collectively, our results suggest carbon nanodots have novel and unique nano-pharmacological properties that can potentially be useful for treating inflammatory disorders such as atherosclerosis.

2708 Carbon-Based Nanomaterials Trigger Inflammasome Activation in Human Macrophages: Role of Toll-Like Receptor Activation and Intracellular Sensing via NLRP3

S. P. Mukherjee[1], F. Andon[1], K. Bhattacharya[1], N. Lozano[4], I. Gessner[3], A. A. Shvedova[2], S. Mathur[3], K. Kostarelos[4], and B. Fadeel[1]. *[1]Karolinska Institutet, Stockholm, Sweden; [2]NIOSH, Morgantown, WV; [3]University of Cologne, Cologne, Germany; and [4]University of Manchester, Manchester, United Kingdom.*

Carbon-based nanomaterials (CBNs) hold great promises in the fields of medicine and engineering due to their intrinsic electro-mechanical properties (Bhattacharya et al. Nanomedicine. 2016;12(2):333-51). However, CBNs may also trigger inflammation leading to potential adverse outcomes. Inflammasomes are key signaling platforms that detect microorganisms as well as sterile stressors, leading to the secretion of pro-inflammatory cytokines, e.g., IL-1β. Here we investigated how different CBNs, including hollow carbon spheres (HCS), single- and multi-walled carbon nanotubes (CNTs), and graphene oxides (GO; with small or large lateral dimensions) are sensed by primary human macrophages which are key cells of the innate immune system. First, we determined the endotoxin content of all the materials, a prerequisite for any studies using immune-competent cells. Then, primary human monocyte-derived macrophages (HMDM) were exposed for 24 h of the various CBNs to assess cytotoxicity. We also performed cytokine profiling using a multiplex assay, and IL-1β secretion was monitored using ELISA, in HMDMs primed or not with lipopolysaccharide (LPS). IL-1β production was noted in presence, but not in the absence of LPS priming, indicative of inflammasome activation. Using THP-1 knockdown cell lines, we found that IL-1β secretion was caspase-1-, ASC-, and NLRP3-dependent. Furthermore, HEK293 reporter cell lines were employed to evaluate the role of Toll-like receptor activation. Notably, SWCNT were apparently sensed by both TLR 2 and TLR4, while GO did not trigger TLR activation. Overall, our study showed that different CBNs, both small, spherical carbon particles (i.e., HCS) as well as small and large GOs, and CNTs, are capable of activating inflammasome, but only when the macrophages are LPS primed.

2709 Inhalation Exposure to Multi-Walled Carbon Nanotubes Reduces T_H2 Immune Responses in a House Dust Mite Model of Asthma

M. D. Ihrie[3], A. J. Taylor[3], G. L. Baker[1], M. D. Stout[2], K. S. Duke[3], and J. C. Bonner[3]. *[1]Battelle, West Jefferson, OH; [2]National Toxicology Program, Research Triangle Park, NC; and [3]North Carolina State University, Raleigh, NC.*

Introduction: Multi-walled carbon nanotubes (MWCNTs) have numerous applications in electronics and engineering. There is increasing evidence that MWCNTs can have harmful effects upon inhalation. Specifically, exposure to MWCNTs may initiate an inflammatory response in the lungs which can influence the development of lung diseases such as asthma, an allergic lung disease characterized by a T_H2 immune response, airway hypersensitivity and mucus cell metaplasia (MCM). Our lab has previously shown that co-exposure of MWCNTs with house dust mite allergen (HDM) causes an enhanced asthmatic phenotype; we therefore hypothesized that inhalation exposure of mice to MWCNTs would render them more susceptible to developing HDM-induced allergic airway disease. Methods: Male B6C3F1/N mice (n=76) were exposed by whole body inhalation for 30 days to 0, 0.06, 0.2 and 0.6 mg/m^3 of 1020 Long MWCNTs (Sun Innovations, Inc.). Mice were then given 25 μg of HDM by intranasal instillation 6 times over 3 weeks. Mice were sacrificed 3 and 30 days after the last HDM dose and bronchoalveolar lavage fluid (BALF), serum and organs were collected. Results: BALF cell counts displayed HDM-induced eosinophilia at 3 days, which decreased by 30 days and was reduced by MWCNT exposure. IL-13 and CCL2 levels were elevated in the BALF of HDM treated mice at 3 days and showed a downward trend with increasing MWCNT dose; IL-1β and TGF-β1 were increased at 30 days by MWCNT treatment. Serum IgE was significantly increased by HDM treatment while MWCNT exposure significantly reduced this effect (p<0.001). Lung histology showed modest airway inflammation caused by HDM or MWCNTs at 3 days, while at 30 days, greater collagen deposition and airway inflammation was caused by MWCNT/HDM exposure. HDM exposure caused MCM, while MWCNT treatment had no significant effect alone nor did MWCNTs alter HDM-induced MCM. Conclusions: Inhalation exposure to MWCNT caused a dose-dependent increase in airway inflammation that was enhanced by HDM exposure. However, MWCNT exposure decreased common allergic markers in HDM-induced asthma, indicating the possibility of differing mechanisms of inflammation. Further work will examine the roles that T_H1 and T_H2 mediated inflammation play in this model. *(Funding: NIEHS grant R01-ES020897)*

2710 Inhibitory Effects of Triamcinolone-Carbon Nanotube Conjugation on Inflammation of Human Arthritis Synovial Fibroblasts

S. Lee[1], and S. H. Kim[2]. *[1]Korea Research Institute of Bioscience and Biotechnology, Jeongeup, Korea, Republic of; and [2]Kyungpook National University, Daegu, Korea, Republic of.*

Repetitive intra-articular corticosteroid injections are inevitable for treating synovial inflammation in advanced arthritis. However, short- and long-term use of corticosteroids usually triggers serious side effects (i.e., adrenal insufficiency, hyperglycemia, Cushing syndrome, osteoporosis, Charcot arthropathy, etc.). This study demonstrated that conjugation of a corticosteroid (triamcinolone) on polyethyleneglycol (PEG)-fabricated multi-walled carbon nanotubes enhances intracellular drug delivery via increased lysosome transport and ultimately suppresses the expression of major pro-inflammatory cytokines (i.e.,TNF-a, IL-1b, and IL-6) and matrix metalloproteinase-1 and -3 from fibroblast-like synoviocytes at a very low drug dose. Specifically, conjugation of triamcinolone and multi-walled carbon nanotubes inactivated nuclear factor-kB via inhibition of the phosphorylation of mitogen-activated protein kinases and the serine/threonine kinase Akt. In summary, low-dose triamcinolone conjugation with carbon nanotubes significantly inhibited the inflammatory response of fibroblast-like synoviocytes by achieving highly efficient intracellular trafficking and suggested a potential drug candidate for resolving side effects associated with conventional arthritis treatment.


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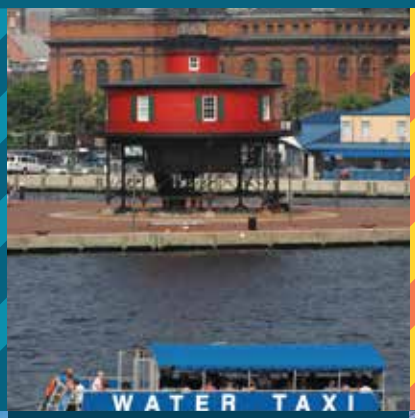

OXFORD
UNIVERSITY PRESS

ISSN 1096-6080
Volume 156, Issue 1
March 2017

www.toxsci.oxfordjournals.org

The Official Journal of the Society of Toxicology

SOT | Society of Toxicology

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