1394 ROLE OF TNFα AND CAVEOLIN-1 IN OZONE-INDUCED INFLAMMATORY MEDIATOR RELEASE AND TOXICITY.

L. Fakhrzadeh, J. D. Laskin and D. L. Laskin. *Environmental and Occupational Health Sciences Institute, Rutgers University/UMDNJ-Robert Wood Johnson Medical School, Piscataway, NJ.*

Exposure to toxic levels of ozone (O_3) cause alveolar epithelial damage. We have previously demonstrated that alveolar macrophages (AM) and inflammatory mediators including nitric oxide generated *via* nitric oxide synthase, PGE2 and TNF-α contribute to the pathogenesis of tissue injury. The generation of these mediators is regulated, in part, by the transcription factor, NF-κB. Our findings that NF-κB p50 knockout mice are unable to generate inflammatory mediators and are protected from O_3 induced lung injury, demonstrate a critical role for this protein in the pathogenic process. In the present studies we analyzed mechanisms regulating NF-κB activation in the lung after O_3 inhalation. Treatment of wild type (WT) mice with ozone (0.8 ppm, 3 h) resulted in a rapid increase in NF-κB binding activity in AM which peaked after 6-12 h. This response was attenuated in mice with a targeted disruption of TNF-α. TNF-α signaling involves p42/44 (ERK 1/2) MAP kinase, and phosphatidylinositol 3'-kinase/protein kinase B (PI3K/PKB) which are important in NF-κB activation and the generation of inflammatory mediators. O_3 inhalation resulted in increased ERK1/2 expression and activation in WT mice which was evident immediately after exposure. Activated ERK 1/2 is known to downregulate caveolin-1 (Cav-1), a negative regulator of PI3K. O_3 inhalation markedly reduced constitutive Cav-1 expression in AM. This was directly correlated with O_3-induced activation of PI3K and PKB. In contrast, in TNF-α-/- mice, O_3 had no effect on Cav-1 or PI3K expression. These data, together with our findings that TNF-α suppresses Cav-1 expression *in vitro*, demonstrate that TNF-α and downstream signaling molecules are important in activation of NF-κB and the regulation of inflammatory genes involved in O_3 toxicity. Supported by NIH grants ES04738, GM34310 and ES05022.

1395 DIFFERENTIAL EXPRESSION OF TREFOIL FACTORS 1 AND 3 FOLLOWING AIRWAY EPITHELIAL CELL INJURY.

G. L. Baker, L. S. Van Winkle, M. V. Fanucchi, D. C. Kim and C. G. Plopper. *Veterinary Medicine: Anatomy Physiology and Cell Biology, UC Davis, Davis, CA.*

Trefoil factors are rapidly induced following mucosal injury in the gastrointestinal tract and function to protect the intestinal mucosa, enhance epithelial cell migration and are essential for epithelial repair in the GI tract. Recently TFF1 and TFF3 have been identified in the human respiratory tract, suggesting that trefoil factors may play a similar role in airway mucosal protection and injury repair processes. The present study was designed to determine whether trefoil peptides are modulated following injury in a well-defined model of airway epithelial cell injury using the Clara cell specific bioactivated pulmonary toxicant naphthalene (NA). Male Swiss Webster mice received an intraperitoneal injection of 200-mg/kg NA dissolved in corn oil carrier. Airways RNA was isolated 0, 1, 3, 6, 12 and 24 hours following naphthalene treatment. Real-time RT-PCR was performed on the isolated RNA to measure airway TFF1 and TFF3 expression. Six hours following naphthalene treatment TFF1 expression was increased 4 fold in the airways and at 24 hours it was increased 7 fold. TFF3 expression was increased 2 fold 1-3 hours following naphthalene treatment and was non-detectable from 6-24 hours following NA treatment. These results demonstrate that TFF1 and TFF3 are differentially modulated following acute airway injury. The increase in TFF3 that occurs immediately following NA administration may represent in an immediate mucosal protective mechanism, while the later TFF1 increase and the loss of TFF3 expression, which first occurs at a time when glutathione depletion occurs, may represent involvement in both airway mucosal protection and repair of injury. Supported in part by NIEHS ES06700, ES04311, ES05707 and the California Tobacco Related Disease Research Program of the University of California Grants TRDRP 121T-0191 and 11RT-0258.

1396 PRE-TREATMENT WITH DIESEL EXHAUST EXTRACT ALTERS INFLUENZA VIRUS REPLICATION IN LUNG EPITHELIAL CELLS.

I. Jaspers[1, 2], J. Ciencewicki[3], M. Beck[2], W. Zhang[1] and M. Brighton[1]. *[1]Center for Env. Med., Asthma, & Lung Biology, University of North Carolina, Chapel Hill, NC, [2]Pediatrics, University of North Carolina, Chapel Hill, NC and [3]Curriculum of Toxicology, University of North Carolina, Chapel Hill, NC.*

Diesel Exhaust (DE) has been demonstrated to generate inflammatory responses in the lung and modify immune responses to inhaled allergens. However, little is known about the effects of DE on common respiratory viral infections. Previous studies have demonstrated that exposure to DE enhances influenza virus replication in mice. We examined whether exposure to DE extracts (DEE) modifies influenza infections of human lung epithelial cells. Differentiated human bronchial epithelial (HBE) cells or A549 cells were exposed to DEE prior to infection with influenza A Bangkok/1/79. At 24 hours - 5 days post-infection total RNA was analyzed for the expression viral genes, as well host cell inflammatory and antiviral genes. Analysis of hemagglutinin (HA) mRNA, a marker for viral propagation, indicates that pre-treatment with DEE enhances viral replication in both differentiated HBE and A549 cells as early as 24 hours post-infection in a DEE dose-dependent manner. This was not caused by a reduced antiviral defense response of the host cells, since influenza-induced mRNA levels for interferon β and MxA, a major interferon β-inducible antiviral defense gene were also enhanced by pre-treatment with DEE. In addition influenza-induced nuclear translocation of phospho-STAT1 and ISGF3γ, transcription factors mediating interferon β-induced gene expression, were also up-regulated by prior exposure to DEE, indicating that interferon β-induced signal transduction pathways are also enhanced by pre-treatment with DEE. Taken together these data indicate that exposure to DE can enhance influenza virus replication in human lung epithelial cells possibly without altering the ability of the host cell to defend itself against the invading pathogen.

1397 SOLUBLE METALS ASSOCIATED WITH ROFA SUPPRESS LUNG IMMUNE DEFENSE AND ALTER CYTOKINE PROFILES AFTER INFECTION IN RATS.

J. R. Roberts[1, 2], M. D. Taylor[1], V. Castranova[1, 2] and J. M. Antonini[1, 2]. *[1]NIOSH, Morgantown, WV and [2]WVU, Morgantown, WV.*

Residual oil fly ash (ROFA), a by-product of fossil fuel combustion and a component of air pollution, has been associated with increased morbidity in susceptible populations. We have shown that soluble metals in ROFA cause lung inflammation and increase susceptibility to infection in rats. The objective was to examine the mechanisms by which soluble metals in ROFA may enhance susceptibility to infection. ROFA was suspended in saline, separated into soluble and insoluble fractions, and the soluble portion (ROFA-SOL) was retained. At day 0, rats were intratracheally instilled (IT) with ROFA (2.0mg/rat) or equivalent quantities of ROFA-SOL, or saline. At day 3, rats were separated into two groups and received an IT dose of either $5x10^4$ or $5x10^5$ *Listeria monocytogenes*. Rats were euthanized on days 3 prior to infection, and on days 6, 8, and 10, bronchoalveolar lavage (BAL) was performed on the right lungs, and bacterial clearance was assessed using the left lung. BAL fluid was centrifuged and the supernatant was retained for measurement of cytokine levels. Exposure to ROFA-SOL significantly decreased pulmonary clearance of bacteria at both doses and decreased animal survival after treatment with the high bacterial dose. Prior to bacteria inoculation on day 3, IL-2 was decreased and IL-6 was increased in rats treated with ROFA-SOL compared to saline. After exposure to both doses of bacteria, IL-2 was decreased whereas IL-6 and IL-10 were elevated in ROFA-SOL-treated rats compared to saline. An elevation in IL-6, a pro-inflammatory cytokine associated with the acute phase response, may partially account for the ROFA-SOL-induced inflammation. An increase in IL-10, a cytokine involved in macrophage inhibition, and a reduction in IL-2, a cytokine promoting T-cell growth and proliferation, may result in a suppression of the innate and adaptive immune response to infection, respectively. ROFA and its associated soluble metals alter cytokine production which may affect the ability of the animals to respond to the infection.

1398 SHORT-TERM EXPOSURE TO INHALED DIESEL EXHAUST PARTICLES ENHANCES ASTHMA-LIKE SYMPTOMS AND INCREASES CYP1A1 mRNA LEVELS.

M. J. Whitekus[1], N. Brechun[2], S. K. Nelson[2], O. Hankinson[1] and D. Diaz-Sanchez[3]. *[1]Department of Pathology and Laboratory Medicine and Jonsson Comprehensive Cancer Center, UCLA, Los Angeles, CA, [2]Webb-Waring Antioxidant Research Institute, Denver, CO and [3]Division of Clinical Immunology and Allergy at UCLA School of Medicine, UCLA, Los Angeles, CA.*

Epidemiological studies suggest a correlation between exposure to ambient particulate matter and adverse health effects in humans, however, there is still a fundamental lack of understanding of the mechanisms involved. We have established a C57BL/6 mouse model to study the adjuvant effects of short-term inhaled diesel exhaust particle (DEP) exposure on asthma. In this model, animals treated with DEP + ovalbumin (OVA) display cellular infiltration in the lung (eosinophila) and demonstrate increased levels of IgG1 in sera compared to ovalbumin treated mice. Our current hypothesis is that DEP mediates some of its adjuvant effects through oxidative stress. However, when we measure the carbonyl content of total mouse lung in our model we found that there was no difference between DEP+OVA and OVA treated mice, suggesting that oxidative stress is not responsible for DEPs modulatory effects on cellular infiltration. Preliminary data using qPCR also suggests that CYP1A1 mRNA levels in total mouse lung increased by 18 fold in DEP+OVA treated mice. Thus, in our mouse model the adjuvant effect of DEP on cellular infiltration might function through another pathway besides oxidative stress, possibly through stimulation of the Ahr. Supported by US Public Health Service Grant AI50495.

Society of Toxicology
43rd Annual Meeting
Baltimore, Maryland

THE TOXICOLOGIST

a supplement to
TOXICOLOGICAL SCIENCES

An Official Journal of the Society of Toxicology

Volume 78, Number S-1, March 2004

Preface

This issue of *The Toxicologist* is devoted to the abstracts of the presentations for the symposium, workshop, roundtable, platform and poster sessions of the 43rd Annual Meeting of the Society of Toxicology, held at the Baltimore Convention Center, Baltimore, Maryland, March 21-25, 2004.

An alphabetical Author Index, cross referencing the corresponding abstract number(s), begins on page 443.

The document also contains a Keyword Index (by subject or chemical) of all the presentations, beginning on page 473.

The abstracts are reproduced as accepted by the Program Committee of the Society of Toxicology and appear in numerical sequence.

Copies of *Program* and CD-ROM with the Itinerary Planner are available at $45 each plus $5 postage and handling (U.S. Funds) from:

Society of Toxicology
1821 Michael Faraday Drive, Suite 300
Reston, VA 20190

http://www.toxicology.org