gene expression in the mouse olfactory mucosa under conditions of regeneration will provide clues to pathways important in neurogenesis and maintenance of olfactory mucosa. We identified highly expressed genes in the olfactory system by mining a gene expression database comparing the expression of 8,700 genes on the Incyte Mouse Gem 1 microarray across 75 developing and adult mouse tissues. The highest expression was noted for genes. Paraoxonase 1 and lactotransferrin were also highly expressed. We segmented the genes into those that were highly expressed in only the olfactory mucosa. One cluster of such regulated genes included sphingosine phosphate lyase,and neuronal leucine-rich repeat protein 1. Because neither the presence nor a role for SPL in the olfactory mucosa has previously been demonstrated, we undertook in situ hybridization analyses to characterize the expression pattern of SPL and other genes identified in this survey. These results revealed highly restricted expression patterns in the olfactory epithelium. We hypothesize that genes such as these may have regulatory roles in olfactory mucosal homeostasis possibly in response to some environmental exposures.

706.7

PCB-induced Inflammatory Reactions in Human Endothelial Cells: Implications in Cancer Metastasis.

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Environmental chemicals, such as polychlorinated biphenyls (PCBs) can markedly increase rates of cancer metastasis. In addition, the direct interaction between tumor cells and adhesion molecules on the surface of vascular endothelial cells is the critical step in the development of metastatic processes. Inflammatory mediators involved in this process include adhesion molecules such as ICAM-1 and VCAM-1, as well as chemotactic factors such as MCP-1. To study the hypothesis that selected PCBs can induce these inflammatory mediators, human umbilical cord endothelial cells (HUVEC) were exposed to increasing doses of PCB 104. A series of RT-PCR analyses revealed that PCB 104 can markedly induce ICAM-1, VCAM-1, and MCP-1 mRNA levels in a dose-dependent manner. The maximum expression of these inflammatory genes was observed in HUVEC exposed to 20 μM of PCB 104. These results indicate that selected PCBs are potent stimulants of inflammatory mediators in human vascular endothelial cells. The possible relevance of this process to the development of cancer metastasis remains yet to be established. Supported by grants from NIEHS).

706.8

EFFECTS OF CIGARETTE SMOKE IN MICE LUNG

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Cigarette smoke is a complex mixture of chemicals countaining more than 4000 differents constituents. There is considerable evidence to suggest that cigarette smoke induced influx of inflammatory cells into the lung is an initial pathogenic factor in the development of emphysema. Emphysema is characterized by abnormal irreversible enlargement of the air spaces in the lung, resulting from progressive degradation of the extracellular matrix of the alveolar walls. Our objective was to investigate histological changes in the extracellular matrix induced by cigarette smoke. Male (n = 18) C57 black mice (20 - 22 g.) were exposed to the whole smoke from nine full commercials cigarettes 3 times/day using an inhalation chamber during 10, 20 and 30 days. Control group (n = 6) were sham-smoked. Stereological methods were used for estimate histophatological changes in lung. Morphological analysis showed an increase in alveolar macrophages number and decreased of the volume density (Vv) of alveolar walls. These data indicate that cigarette smoke induced changes in the septal extracellular matrix and suggest macrophages have the critical role of lung injury. Supported by: FAPERJ, CAPES and CNPQ.

706.9

Effects of Manganese on the Estrus Cycle of Sprague Dawley Rats

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The aim of the present study was to investigate if manganese exposure caused any disturbances in the estrus cycle of the female rats. The vaginal test allows one to determine the extent to which normal function may be disrupted by toxicant exposure. Rats (n=40) were grouped in to five groups of 8 rats each. Vaginal smears were collected for 10 days before the dosing started. Knowledge of the vaginal smear prior to treatment provides a point of comparison immediately after the initiation of treatment and for any subsequent changes that may ensue. The groups were designated as control (0) mg/kg, control positive (0) mg/kg, A (76.5) mg/kg, B (150.0) mg/kg and C (306) mg/kg. The rats were dosed every morning between 10:00 and 12:00 by oral gavage for a period of 63 days with manganese acetate. Control and control positive groups received water. After collecting smears every morning, they were fixed in alcohol, stained with H & E. studied microscopically for quantification of epithelial cells. Frequency of cornified cells was calculated as a percentage. The interval in days between the successive peaks in the frequency of cornified cells as the length of individual estrus cycle. Preliminary analysis of our results revealed that manganese treatment did not significantly alter the length of the estrus cycle in female rats.

706.10

Mechanisms for arsenite-induced angiogenesis.

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Chronic, low-level exposure to arsenite increases the incidence of proliferative vascular diseases. Arsenic-stimulated angiogenesis may contribute to the vascular components of hypertension, diabetes and tumor growth. In support of this hypothesis, arsenite (0.033-1.0 μM) stimulated angiogenesis in a chicken embryo model. Above 1.0 μM, arsenite inhibited angiogenesis. Arsenite increased expression of the angiogenic genes, vascular endothelial cell growth factor (VEGF) and plasminogen activator inhibitor-1 (PAI-1), in porcine aortic smooth muscle cell (SMC) cultures. Release of VEGF protein increased following a 24 h exposure to 5-25 μM arsenite. By 48 h, 1 μM arsenite caused significant VEGF release. Changes in VEGF mRNA levels were consistent with the changes in protein levels. In contrast to VEGF, PAI-1 mRNA levels were stimulated by low levels of arsenite, but inhibited by exposures above 2.5 μM arsenite. Low arsenite levels also increased hypoxia inducible factor (HIF) protein levels in a manner consistent with enhancement of VEGF and PAI-1 promoter activity. Inhibitor studies demonstrated that arsenite stimulated oxidant formation and kinase cascades to increase HIF levels and angiogenic gene expression. These data suggest that arsenite causes specific, dose- and time dependent effects on cell signaling that promote angiogenic responses, which contribute to pathologic vascular changes. Supported by ES07373.

ENVIRONMENTAL LUNG INJURY (707.1-707.8)

707.1

Influence of hyperthyroidism on rat lung cytokine production and nuclear factor (NF)-kappa B activation following ozone exposure.

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Results from previous studies indicate that hyperthyroidism increases the risk of ozone-induced lung toxicity. In the present study, we evaluated cytokine levels in bronchoalveolar lavage fluid from control and hyperthyroid male rats 18 hours after exposure to air or ozone (2 ppm for 3 hours). MIP-2 and MCP-1 levels were increased in both control and hyperthyroid rats following ozone. However, the increases in hyperthyroid rats were much greater, MIP-2 (1.5 fold) and MCP-1 (11 fold), when compared to levels in controls. These changes appeared to be relatively specific; bronchoalveolar lavage fluid levels of IL-6, IL-4, and IL-10 were generally low or non-detectable across all groups at this time. We also found that NF-kappa B binding activity was increased at both 4 and 18 hours following ozone exposure in bronchoalveolar lavage cell extracts from hyperthyroid rats relative to controls. Collectively, these results suggest that mechanisms contributing to the enhanced pulmonary

inflammatory response to ozone in a hyperthyroid state include an increase in NF-kappa B activation and an up-regulation of chemokine production.

707.2

Induction of CYP1A1 and NADPH quinone oxidoreductase (QR) with concomitant attenuation of CYP2B1 in the lung: Effects of paving asphalt fume exposure

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Polycyclic aromatic hydrocarbons (PAHs) in asphalt fumes require bioactivation by cytochrome P-450 to exert toxic/carcinogenic effects. Exposure to asphalt fume may alter phase I (P-450) and phase II (QR and GST) enzymes in the lung. The objective of this study was to evaluate this hypothesis in a rat model. Rats were exposed to air or asphalt fume generated at paving temperature (~150°C, ~20 mg/m^3, 3.5h or 6h/day for 5 days). Lung microsomes and cytosol were isolated. The activities for P-450 isozymes (CYP1A1 and CYP2B1) and phase II enzymes (QR and GST) were monitored. Protein levels were determined by Western blot analysis. The results show that asphalt fume exposure significantly induced the activity and protein levels of CYP1A1, but markedly reduced CYP2B1 levels and activity in the lung. By immunofluorescence, CYP1A1 induction was localized in airways, blood vessels, and alveoli. Cytosolic QR activity was significantly elevated after asphalt fume exposure, whereas, GST activity was not affected by the exposure. This induction of CYP1A1 and QR with the concomitant attenuation of CYP2B1 after asphalt fume exposure may alter PAH metabolism leading to potential carcinogenic/mutagenic effects.

707.3

Alternation of Innate and Cell-Mediated Immunity to Listeria monocytogenes by Short-Term Exposure to Diesel Exhaust Particles

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The effects of diesel exhaust particles (DEP) on functions of pulmonary host defense were studied using a rat Listeria infection model. Short-term DEP inhalation (50 and 100 mg/m3, 4 h) by rats resulted in a slowed lung bacterial clearance, suppressed alveolar macrophage (AM) phagocytosis and reduced AM production of tumor necrosis factor (TNF)-alpha, interleukin (IL)-1 and IL-12 in response to Listeria. The combined DEP/Listeria exposure was also characterized by increased CD4+ and CD8+ cell counts, and percentage of CD8+ cells in lung draining lymph nodes. The lymphocytes from DEP-treated rats showed increased IL-2 production when challenged with concanavalin A (ConA). Cells from the combined exposed rats produced increased interferon (IFN)-gamma at day 7, but decreased IFN-gamma at day 3, when challenged with either ConA or heat-killed Listeria (HKLM). Listeria significantly induced lymphocyte secretion of IL-6 in response to HKLM, which could be increased by DEP preexposure. These results indicate that short-term DEP exposure aggravates Listeria infection by suppressing AM phagocytosis and the production of TNF-alpha, IL-1 and IL-12, and by eliciting an adverse effect on T-cell-mediated immunity.

707.4

Effects of paving asphalt fume exposure on genotoxic and mutagenic activities in the lung

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Asphalt fumes are complex mixtures of aerosols and vapors containing various organic compounds, including polycyclic aromatic hydrocarbons, and have been demonstrated to alter the cytochrome P-450 system in the lung with significant induction of CYP1A1. The present study was carried out to characterize the potential mutagenic and genotoxic effects of inhaled asphalt fume in a rat model. Rats were exposed to air or asphalt fume generated at paving temperature (~150°C) at ~98 mg/m^3 and 6h/day for 5 days. Alveolar macrophages (AM) were obtained by bronchoalveolar lavage. S9 was isolated from the lung tissue. Mutagenicity was monitored using the Ames test and genotoxicity was determined by Comet assay. In comparison to controls, AM from asphalt fume-exposed rats showed significant DNA damage as demonstrated by the Comet assay. However, the S9 fraction from asphalt exposed rat lungs used for metabolic activation of 2-aminoanthracene or benzo[a]pyrene in Ames tests, did not induce mutagenic activity in either *S. typhimurium* YG1024 or YG1029. These results suggest that short term inhalation of asphalt fume did not induce S9 metabolic activity-dependent mutagenicity, but caused significant DNA damage in AM.

707.5

Induction of Apoptosis from Instillation of Agricultural Dust

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In this study, we test the hypothesis that induction of alveolar macrophage (AM) apoptosis plays a role in lung injury from particulates. We examined AM apoptosis in F344 rats following intratracheal instillation of 1ml of saline containing 4mg of particles. Particles used included crystalline silica, titanium dioxide and particulates from air sampling of citrus and grape fields. After 4 weeks, rats were sacrificed and lung sections analyzed for the number of apoptotic cells per lung using TdT labelling. Apoptotic macrophages, expressed in millions of cells per lung (mean±SE,N=4), were 43.4±4 for crystalline silica, 10.50±0.2 for grape, 8.1±0.5 for citrus and 3.1±0.4 for titanium dioxide. Saline controls were 1.6±0.4. The degree of apoptosis correlated well with inflammation and damage suggesting a role for apoptosis in lung injury resulting from particulates.

707.6

Enhanced Nitric Oxide Production Associated with Silica-Induced Disease in Rats.

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This study investigated the relationship between silica-induced lung damage, inflammation and fibrosis and nitric oxide (NO) production in rats exposed by inhalation to silica (15 mg/m^3 silica, 6 hr/day, 5 days/week, for 116 exposure days). Markers of pulmonary inflammation and damage rise rapidly (5-16 days of exposure) and are maintained at a relatively stable new set point through 40 days, before rising explosively at 79 and 116 days of exposure. Fibrosis is evident only during this explosive period of inflammation and damage. Silica inhalation results in a rapid (5 days) rise in NO-dependent chemiluminescence from alveolar macrophages (AM). NO products in bronchoalveolar lavage fluid increase significantly, exhibiting a time course similar to that for parameters of inflammation and damage. Immunohistochemical analysis of pulmonary inducible NO synthase (iNOS) and nitrotyrosine identified elevated iNOS and NO-induced damage in AM and type II cells after 79 days of exposure. This NO production is associated with granulomatous regions of the lung. These data indicate that NO production is associated temporally and anatomically with silica-induced lung disease in a rat inhalation model.

707.7

Increased lung permeability accompanies homeostatic decline in aging mice

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Epidemiological studies show elderly populations are at greater risk for adverse health effects from inhaled environmental particulate matter. To determine whether enhanced particle uptake contributes to increased risk, we studied 99mTc-DTPA clearance from the lung in mice with genetically accelerated aging(AKR/J). Onset of weight loss in this strain predicts lifespan and is a reliable estimate of homeostatic decline(34±3SE)(Tankersley, AJP 2001). Both stable adult(wild type:25g, AKR/J:45g) and AKR/J mice after 9.3% weight loss were anesthetized, intubated, and given 25µl 99mTc-DTPA by tracheal instillation. 30min clearance from the lung was measured by gamma scintigraphy. Results indicated that mice showed an increased rate of particle clearance after weight loss compared to stable adult controls. Particle retention in the lung of stable adult wild type(n=4), AKR/J(n=2), and senescent AKR/J(n=8) averaged 56%, 50% and 40%($p<0.05$), respectively. Thus during the period of homeostatic decline

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ABSTRACTS

(continued from Part I)

MONDAY MORNING
April 22, 2002

American Association of Anatomists

MONDAY AFTERNOON
April 22, 2002

American Society for Biochemistry and Molecular Biology

American Society for Nutritional Sciences

American Association of Anatomists

TUESDAY MORNING
April 23, 2002

Experimental Biology

The American Physiological Society