ENDOTOXIN-RELATED INFLAMMATORY RESPONSE IN NASAL LAVAGE FLUID AFTER NASAL PROVOCATION WITH SWINE CONFINEMENT DUSTS

D. Nowak[1], G. Denk[2], R. Jörres[2], D. Kirsten[2], F. Koops[1], D. Szadkowski[1], B. Wiegand[3], J. Hartung[4], H. Magnussen[2]

[1]Zentralinstitut/Ordinariat für Arbeitsmedizin, 22083 Hamburg, FRG, [2]Krankenhaus Grosshansdorf, FRG, [3]Tierärztliche Hochschule Hannover, FRG, [4]Silsoe Research Institute, GB.

Among pig farmers, high prevalences of respiratory symptoms and lung function impairment have been reported. Bronchoalveolar lavage (BAL) studies indicate an inflammatory cell response among swine confinement workers (Chest 101:767,1992). The aim of our study was to analyze the inflammatory response in nasal lavage (NAL) after nasal challenges with dusts from swine confinement houses. Endotoxin was quantified by a Limulus Amebocyte Lysate test. Three concentrations of endotoxin (2,26,196 µg/g dust) were chosen. On four different days, 9 healthy volunteers (mean age 28±4 yrs) without prior exposure to animal house dust were challenged, in random order, with 50 mg of dust, or sham, administered into each nostril. NAL, spirometry and acoustic rhinometry were performed immediately before, 60, and 180 min after each exposure. Cells were counted by haemocytometer, differential counts were done on cytocentrifuge preparations, and albumin was measured by RIA. Mean±SD values for total cells (x10^3/ml), % neutrophils and albumin (ng/l) are given in the table:

Endotoxin content		0 µg/g	2 µg/g	26 µg/g	196 µg/g
Cells	0 min	4.9±4.5	6.2±6.1	5.1±3.4	4.5±3.3
	60 min	3.1±2.0	4.3±4.1	7.9±8.2	2.8±1.7
	180 min	4.1±4.1	58.5±67.0	88.0±139.9	91.0±139.3
% neutro	0 min	37±33	42±21	34±25	18±22
	60 min	31±31	39±20	41±27	31±26
	180 min	40±26	90±5	77±30	90±8
Albumin	0 min	8.9±11.1	7.4±4.5	8.2±14.0	7.6±8.5
	60 min	4.8±4.9	5.7±4.9	9.7±8.0	4.2±3.4
	180 min	6.6±7.8	12.8±9.9	13.5±17.8	14.7±24.5

For cell numbers, % neutrophils and albumin, ANOVA revealed significant ($p<0.05$) increases 180 but not 60 min after administration of endotoxin, with no significant dose-dependence. Spirometry and acoustic rhinometry did not demonstrate any changes.

Our findings suggest that (1) inflammatory changes in NAL fluid after endotoxin administration from swine confinement houses resemble those reported for BAL fluid, and (2) within the range analyzed, the lowest dose of endotoxin administered (0.2 µg) already causes an almost maximal inflammatory response.

MUCOUS CELL METAPLASIA (MCM) IN THE RESPIRATORY TRACT OF RATS AFTER EXPOSURE TO COTTON DUST AND MACHINING FLUID.

Terry Gordon and Jack Harkema. Institute of Environmental Medicine, NYU Medical Center, Tuxedo, NY, USA; and Inhalation Toxicology Research Institute, Albuquerque, NM, USA.

Increased sputum production and chronic bronchitis are associated with occupational exposure to bio-aerosols contaminated with endotoxin. The purpose of the present study was to determine if occupationally relevant concentrations of cotton dust and machining fluids can induce MCM in rat airways. F344 rats were exposed to: 1) filtered air or 1.5, 5.0, or 15.0 mg/m^3 cotton dust for 2 h/d; or 2) aerosols of water or 1 or 10 mg/m^3 machining fluids for 3h/d for 3 d and sacrificed at 24 h after the last exposure. Tissues from the proximal nasal septum and axial airway of the left lung lobe were processed and stained with AB/PAS to identify stored mucosubstances in the epithelium (M_E). Significant increases in M_E were evident in the nasal septum of rats exposed to 5.0 and 15.0 mg/m^3 cotton dust (5.6 and 3.5 fold greater, respectively). Smaller, yet statistically significant increases in M_E were also observed in the axial airways of these 2 groups of animals (1.6 and 2.2 fold greater, respectively). No changes in the amounts of M_E were observed in the respiratory tract of rats exposed to 1.5 mg/m^3 cotton dust compared to air controls. Similar to the MCM induced by cotton dust, exposure to 10 mg/m^3 used machining fluids produced significant increases in M_E in the nasal septum and axial airway (5-fold and 1-fold greater, respectively). Exposure to 10 mg/m^3 unused machining fluids also induced a significant increase in M_E in the nasal septum (4-fold), but not in the axial airway. No changes in the amounts of M_E were observed in the respiratory tract of rats exposed to 1.0 mg/m^3 used machining fluid. These studies suggest that while contaminating endotoxin may play a significant role in the increase in sputum and chronic bronchitis reported for workers exposed to organic dust and aerosols, other irritant factors in machining fluids participate in the induction of MCM in the respiratory epithelium of the nasal septum.

(Supported by NIH ES04947, NIOSH 03044, and US DOE/OHER Contract No. DE-AC04-76EV01013.)

RAPID PRIMING OF MONOCYTE TNF-α RELEASE IN WORKERS EXPOSED TO GRAIN DUST.

Paul JA Borm[1], Roel P.F. Schins[1], Thim JJM Derhaag[1], IJmert Kant[2] Tim HJM Jorna[3]. [1]Department of Health Risk Analysis and Toxicology; [2]Department of Epidemiology; [3]Department of Respiratory Diseases, University of Limburg, Maastricht, The Netherlands .

Endotoxin is considered to be a main causative component in acute and chronic respiratory effects of grain dust exposure. Since cytokines, such as tumor necrosis factor-α (TNF), are involved in these processes, we investigated changes in blood monocyte TNF release in grain dust-exposed workers during one week in relation to individual exposure and lung function. In 12 workers in animal feed industry, lung function was measured daily before and after shift, using flow-volume curves and impedance. On Monday and Friday blood samples (30 ml) were drawn and monocytes were isolated by buoyant density centrifugation / culture dish adherence. Spontaneous TNF release, and TNF release upon the *ex-vivo* stimulation with 3 ng/ml or 1000 ng/ml endotoxin was measured after 18h of incubation in cell free supernatant by ELISA. Individual exposures were determined from stationairy dust measurements at every workplace combined with personal task analysis during all shifts. Variations in lung function parameters were sometimes observed but not related to daily exposure, or to cumulative exposure. The spontaneous TNF release on Friday was on average 6-fold higher compared to Monday (paired t-test, $p<0.001$). The capacity of monocytes to release TNF remained unchanged as no difference was observed between Monday and Friday TNF release upon ex-vivo stimulation with 1000 ng/ml endotoxin. Remarkably, the individual cross-shift (=week) increase in spontaneous TNF release was significantly related ($n=11$, $r=0.75$, $p<0.02$) to the calculated individuals' respirable grain dust exposure. Our data suggest a rapid, dose dependent priming of blood monocytes upon grain dust inhalation. Currently, we're investigating the role of endotoxin as a causal agent and the mediators involved in this rapid priming.

IN VITRO PHARMACOLOGIC STUDIES OF POULTRY DUST EXTRACT.

E. Neil Schachter, Eugenia Zuskin, Nicholas Rienzi, James Godbold, Mount Sinai Medical Center, New York, N.Y, USA; Andrija Stampar School of Public Health, Zagreb, Croatia.

Supported in part by NIOSH RO1-OHO-2593-01A1.

We have previously used guinea pig tracheal (GPT) rings to analyze the effect of organic dusts on airway smooth muscle contraction. In a recent study we documented that workers exposed to dust in the poultry industry develop a high frequency of respiratory findings. A water soluble extract of dust obtained from poultry confinement buildings (PDE) was prepared as a 1:10 w/v solution by the method of Sheldon. Dose related contractions of nonsensitized GPT were demonstrated using the extract. PDE was added to the GPT's in 1/2 log increments in volumes of 10,30,100,300, and 1000 µl. A series of 12 organ baths were used simultaneously. Each GPT was divided into 4 segments allowing us to examine three pharmacologic agents (and one control) in 3 animals at once. The effects of three mediator modifying drugs, indomethacin (10^{-6}M), pyrilamine (10^{-6}M) and LY171883 (10^{-6}M) were studied as well as the effects of two enzyme inhibitors captopril (10^{-5}M) and thiorphan (10^{-5}M). Twelve animals were studied for each agent. Significant reductions in PDE induced contractions were demonstrated in GPT pretreated with all three mediator modifying agents. Similarly both enzyme inhibitors reduced PDE's effect on GPT. We conclude that 1) PDE causes dose related contractile responses in GPT, 2) Mediator modifying agents reduce PDE's contractile effect which is presumably the result of mediator release and 3) Enzyme inhibitors also reduce PDE's contractile effect by an as yet unexplained mechanism. Supported in part by NIOSH grant RO1-OHO-2593-01A.

	Control	I	P	L	C	T*
Emax+	111%	75%	84%	79%	59%	56%

* I = indomethacin; P = pyrilamine; L = LY17188; C = captopril; T - thiorphan

+ Emax as a percent of maximal carbachol contraction.

AMERICAN JOURNAL OF

Respiratory and Critical Care Medicine

Formerly the American Review of Respiratory Disease

ISSN-1073-449X

SUPPLEMENT

April 1994 Volume 149 Number 4, Part 2

AMERICAN LUNG ASSOCIATION • AMERICAN THORACIC SOCIETY

ABSTRACTS

1994 International Conference

May 21–25, 1994 • Boston, Massachusetts

This special supplement of the *American Journal of Respiratory and Critical Care Medicine* contains abstracts of the scientific papers to be presented at the 1994 International Conference, which is sponsored by the American Lung Association and the American Thoracic Society. The abstracts appear in order of presentation, from Sunday, May 22 through Wednesday, May 25 and are identified by session code numbers. To assist in planning a personal schedule at the Conference, the time and place of each presentation is also provided.