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Toxicology of Ammonium Sulfate in the Lung

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There has been some concern that the use of the platinum/palladium (Pt/Pd) catalytic converter to reduce automobile engine exhaust pollutants would result in an increase in ambient sulfuric acid levels. This concern was increased by the finding that sulfuric acid is emitted from the exhaust of the Pt/Pd catalyst equipped cars in a very fine mist with particles ranging from 0.01 to 0.1 μm diameter (U. S. EPA 1978). The toxicity of the sulfuric acid could be enhanced by the large number of particles at a given concentration and effective penetration into the deep lung due to their small size (Task Force on Lung Dynamics 1966).

Most sulfur in ambient air, however, exists in the form of sulfate, primarily ammonium sulfate (STEVENS & DZUBAY 1977). Moreover, there is little evidence that the concentration of the free acid is increasing significantly. Evidently, much of the sulfuric acid produced is neutralized fairly quickly. Furthermore, LARSON et al. (1977) have shown that sufficient ammonia is present in the respiratory tract (up to 520 $\mu g/m^3$) to neutralize probable ambient levels of sulfuric acid. Thus, ammonium sulfate appears to be the primary sulfur compound actually present in the lung from inhalation of ambient acid sulfur pollutants.

Limited evidence indicates that ammonium sulfate is relatively non-toxic. AMDUR (1970) and SIM & PATTLE (1957) have shown that ammonium sulfate is less irritating to the guinea pig lung than sulfuric acid. LIPPMAN (1977) reported no change in pulmonary flow resistance or compliance in donkeys exposed for one hour to more than 4.3 mg/m^3 ammonium sulfate. LOSCUTOFF (1977) found only minor pulmonary function changes in dogs exposed to sulfuric acid until the exposure concentration was sufficient (6.3 mg/m^3) for some unneutralized sulfuric acid to be exhaled. One positive finding was a decrease in ventilatory volume in perfused guinea pig lungs during inhalation of ammonium sulfate, an effect that could be mimicked by histamine or blocked by an H-1 antihistamine (CHARLES & MENZEL 1977).

Most inhalation studies of ammonium sulfate to date have involved acute exposure with emphasis upon measurement of airway constriction. The present study was designed to evaluate the effects of acute, and subchronic exposure to very high concentrations upon arterial blood gases, lung volumes and pathology.

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METHODS

Animals were exposed to ammonium sulfate aerosols in 0.31-m^3 (11-ft^3) stainless steel chambers. The animals were housed in 28 cm (11 in.) square wire cages, two animals per cage. Food and water were provided ad libitum. Ammonium sulfate aerosol was generated from an aqueous solution with either one or two Retec nebulizers (Retec Development Laboratory, Portland, Oregon) and dried by mixing with dry air and passing it through a heated glass tube. Two nebulizers were used at concentrations greater than 500 mg/m^3. Ammonium sulfate concentration was determined by collecting the aerosol on a Gelman A- E glass fiber filter at a flow rate of 2 l/min for 15 min and weighing the filter. Accuracy of the method was periodically checked by chemical analysis of the filter sample. Particle size was evaluated gravimetrically using an Andersen multi-stage sampler (Andersen Sampler Inc., Atlanta, Georgia).

Preliminary exposures were conducted at the maximum attainable concentration of ammonium sulfate using equipment and methods available. A group of six young adult male rats were exposed 8 h/day for 3 consecutive days to a concentration of 1000-1200 mg/m^3. Guinea pigs were also exposed for a single 8 h period. Twenty were exposed to a concentration of 800-900 mg/m^3, six to a concentration of 600-700 mg/m^3 and six to a concentration of 500-600 mg/m^3. The animals were observed for mortality and signs of gross toxicity.

Groups of ten adult male rats were exposed 8 h/day for 1, 3, 7 or 14 days at a concentration of about 300 mg/m^3. After exposure, arterial blood samples were collected from the fully conscious animals and analyzed for blood gases, pH, and standard bicarbonate, using methods previously described (PEPELKO & DIXON 1975). Residual volume (RV) and vital capacity (VC) were determined in rats exposed for 14 days. RV was estimated by helium dilution of an injected gas bolus following a maximal expiration. VC was estimated by filling the lungs to a pressure of 30 cm H_2O from residual volume.

Immediately after blood and lung volume measurements, the animals were sacrificed and the lungs fixed in 10% formalin by intratracheal instillation at 30 cm H_2O pressure. Paraffin sections were stained with hematoxylin eosin for microscopic observation.

RESULTS AND DISCUSSION

At a concentration of 1000-1200 mg/m^3, particle size averaged approximately 2-3 μm diameter. At lower concentrations, the mean diameter was slightly less, generally ranging from 1-2 μm diameter at 300 mg/m^3. Concentrations of ammonium sulfate during subchronic exposures were maintained within $\pm$ 10% of the desired concentration.

In the preliminary exposures, none of the rats died during three consecutive days of exposure to 1000-1200 mg/m^3 ammonium sulfate. No gross toxicological effects were noted. Eight of 20 guinea pigs exposed to 800-900 mg/m^3, 1/6 exposed to 600-700 mg/m^3 and 0/6 exposed to 500-600 mg/m^3 died during exposure. The animals dying during exposure appeared to do so as a result of acute shock and airway constriction. Any sudden noise or other disturbance was likely to precipitate such an event. After exposure, the survivors recovered with no noticeable after effects.

Arterial blood gases, pH, and bicarbonate of rats exposed 1, 3, 7 or 14 days are shown in Table 1. No significant differences could be detected between control and exposed rats for any of the parameters measured. Body weights, VC, RV and wet lung weights of rats exposed 14 days to 300 mg/m^3 ammonium sulfate are shown in Table 2. Again, none of these parameters were significantly affected by exposure. Histological examination of the trachea, bronchial lymph nodes and lungs revealed no changes that could be definitely attributed to exposure.

In the present study, exposure of rats to concentrations of ammonium sulfate hundreds of times greater than would be expected to occur in polluted air did not result in detectable toxicological effects. Mortality could not be induced in rats even at concentrations of 1000-1200 mg/m^3. At this concentration, visibility in the chamber was definitely decreased and ammonium sulfate was rapidly deposited upon all surfaces. While mortality did occur in guinea pigs, this was apparently the result of acute airway constriction resulting in asphyxiation and not due to gross lung damage.

As a result of these findings, it was concluded that inhaled ammonium sulfate is relatively non-toxic except at very high concentrations. It was also concluded that any concentration of sulfuric acid likely to be present in the atmosphere is probably non-toxic since the chances of neutralization to ammonium sulfate in the respiratory tract is very great. For example, up to 520 μg/m^3 ammonia has been found in the human respiratory passages

TABLE I

Arterial Blood Measurements After 1, 3, 7, 14 Days
Exposure to 300 mg/m^3 Ammonium Sulfate

Days Exposure	Arterial Po_2 (Torr)		Arterial PCo_2 (Torr)	
	Control	Exposed	Control	Exposed
1	94 ± 4	96 ± 6	37 ± 3	36 ± 4
3	96 ± 2	91 ± 5	36 ± 2	36 ± 3
7	93 ± 3	96 ± 7	34 ± 4	33 ± 4
14	89 ± 5	94 ± 8	37 ± 4	36 ± 3

Days Exposure	Arterial pH		Standard Bicarbonate (MM/L)	
1	7.47 ± 0.03	7.45 ± 0.04	28.6 ± 2.4	27.6 ± 4.4
3	7.46 ± 0.02	7.46 ± 0.03	26.6 ± 0.8	28.6 ± 3.1
7	7.47 ± 0.02	7.49 ± 0.04	27.4 ± 1.1	26.5 ± 1.6
14	7.47 ± 0.01	7.45 ± 0.03	27.4 ± 2.6	26.7 ± 0.8

* Standard Deviation
** N = 10 For All Groups

TABLE 2

Repeated Body Weight Measurements, Vital Capacity
Residual Volume and Wet Lung Weights of Rats Exposed
14 Days to 300 mg/m^3 Ammonium Sulfate

		CONTROL	EXPOSED
Body Weight Pre-Exposure	(GMS)	435 ± 20	447 ± 17
Body Weight 1 Day	(GMS)	420 ± 15	437 ± 21
Body Weight 3 Days	(GMS)	432 ± 17	441 ± 20
Body Weight 7 Days	(GMS)	442 ± 20	445 ± 18
Body Weight 14 Days	(GMS)	457 ± 22.0	446 ± 17
Vital Capacity	(CC)	17.1 ± 2.0	16.6 ± 1.9
Residual Volume	(CC)	1.92 ± 0.26	1.75 ± 0.26
Wet Lung Weight	(GMS)	2.12 ± 0.41	2.11 ± 0.17

* Standard Deviation
* N = For All Groups

(LARSON et al. 1977), while the maximum 24 h mean sulfate concentration for selected cities from 1957-1964 was only 95 $\mu g/m^3$ (MORGAN et al. 1970), with only a fraction of this being free sulfuric acid. The likelihood of neutralization is enhanced further by the fact that ammonia can react with greater than five times its own weight of sulfuric acid.

Despite the relatively low toxicity of ammonium sulfate in experimental animals, it cannot be concluded that increased sulfuric acid production is harmless to human health. Many other pollutants are present in ambient air with possible synergistic effects. Sulfuric acid undoubtedly reacts to produce other sulfates in ambient air which are often much more toxic. For example zinc sulfate and zinc ammonium sulfate are much more irritating to the lung than ammonium sulfate (AMDUR & CORN 1963). In order to assess with more certainty, the health effects of increased sulfuric acid production, it will be necessary to determine accurately that proportion inhaled as free sulfuric acid compared with ammonium sulfate as well as the proportion and kinds of other sulfates present in the atmosphere.

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REFERENCES

AMDUR, M.O.: Proc. Am. Philos. Soc. 14, 3 (1970.
AMDUR, M.O., and M. CORN: Am. Ind. Hyg. Assoc. J. 24, 326 (1963).
CHARLES, J.M., and D.B. MENZEL: Proceedings Catalyst Research Program Sulfuric Acid Research Review Conference. Research Triangle Park, North Carolina: U.S. Environmental Protection Agency 1977.
LARSON, T.V., COVERT, D.S., FRANK, R., and R.J. CHARLSON: Science 197, 161 (1977).
LIPMANN, M: Proceedings Catalyst Research Program Sulfuric Acid Research Review Conference. Research Triangle Park, North Carolina: U.S. Environmental Protection Agency 1977.
LOSCUTOFF, S.M., BURTON, F.B., and B.W. KILLARD: Fed. Proc. 37, 867 (1978).
MORGAN, G.B., OZOLINS, G., and E.C. TABOR: Science 170, 289 (1970).
PEPELKO, W.E., and G.A. DIXON: J. Appl. Physiol. 38, 581 (1975).
SIM, V., and R. PATTLE: J. Amer. Med. Assoc. 165, 1908 (1957).
STEVENS, R., and T. DZUBAY: Proceedings Catalyst Research Program Sulfuric Acid Research Review Conference. Research Triangle Park, North Carolina: U.S. Environmental Protection Agency 1977.
Task Group on Lung Dynamics: Health Phys. 12, 173 (1966).
U.S. Environmental Protection Agency. Workshop on Health Effects of Transportation RElated Pollutants. EPA-6001/1-78-011. Research Triangle Park, North Carolina: EPA 1978.