Meetings

Air Pollution and Medical Research

The effect of air pollution on human welfare was the main theme of the sixth annual air-pollution medical research conference which was sponsored by the California State Department of Public Health, and held in San Francisco (28–29 January). The conferences are intended to encourage communication among scientists involved in research on air pollution and to inform officials who are concerned with air-pollution control and public health about current research. The meetings were organized into three major sessions: trends in air pollution and its effects; the body burden of pollutants, its estimation and interpretation; and laboratory and epidemiological relationships of respiratory disease and pollutant exposures. In addition, a special lecture on public policy in air conservation was given by James P. Dixon, chairman of the Air Conservation Commission of the American Association for the Advancement of Science.

Emission trends of hydrocarbons in Los Angeles County have gone downward for the petroleum industry but upward for transportation sources (Kanter and Lunche, Los Angeles County Air Pollution Control District). Reductions in sulfur dioxide emissions were brought about by constraints on fuel oil used for power-generating plants. There was little evidence of long-range change in average levels of atmospheric particulates during the past 5 years. Sulfur dioxide appears to have decreased from the average values of about 0.04 to 0.02 part per million, with a random variation. In striking contrast to sulfur dioxide levels was a rising trend for carbon monoxide from average values of 6 ppm in 1956 to about 11 ppm in 1961, thus reflecting the excess of motor vehicle emissions over the effects of industrial controls. Oxidant levels, which best reflect the intensity of photochemical smog, show small, if any, decrease. Nitrogen dioxide levels are relatively unchanged over the years, but nitric oxide has been increasing. As a result, the combined oxides have increased from average values of 0.08 to 0.18 ppm in downtown Los Angeles.

Reports on pollution outside Los Angeles County show that oxidant levels in Fresno in 1961 exceeded, more than any other location in northern California, the state's "adverse" standard on 52 days. In Sacramento the "serious" standard for carbon monoxide (average 30 ppm for 8 hours) was exceeded on 7 days with peak values rivaling the highest values observed in the Los Angeles Basin (Taylor, California State Department of Public Health).

Middleton (University of California, Riverside) reported that vegetation damage from air pollutants was gradually increasing, until it now occurs throughout the Los Angeles and San Francisco Bay area air sheds. It is steadily spreading in the Central Valley air shed.

Breslow (California State Department of Public Health) exhibited great concern over the possible relationship of air pollution to chronic diseases, especially of the lung. Establishment of scientifically valid air-quality standards has guided recent air-pollution health research toward more emphasis on physiological changes such as impairment of lung function and interference with oxygen transport by the lung. However, work continues on mortality studies.

Atmospheric reaction investigations have revealed the irrelevancy of measuring "total hydrocarbons" which include methane and certain other hydrocarbons that are essentially unreactive in photochemical systems (Altshuller, Laboratory of Engineering and Physical Sciences, Division of Air Pollution, Public Health Service, Cincinnati). Defects also exist in such studies with synthetic atmospheres because they fail to produce biological and physical effects, for example eye irritation, encountered in natural atmospheres. Much current research effort is devoted to photochemical studies of the rates and sequences of reaction of various compounds, such as aldehydes, aromatic hydrocarbons, and mixtures of organic substances. New analytic tools are playing an increasingly important role.

Goldsmith (California State Department of Public Health) opened the second session on the body burden of pollutants by calling attention to the importance of the concept in setting air-quality standards. A classification of pollutants according to their effects was presented.

Several mathematical models for lead uptake and excretion were presented by Sterling *et al.* (Kettering Laboratory, University of Cincinnati). They also noted the relationship which such models have to the body burden of lead. Data on trace metal levels in human blood and serum taken from three different groups of subjects were shown to be similar for most of the 15 metals used. Slightly more lead was present in the serum of automobile drivers who drove over 500 miles a month than in those who drove less (Butt *et al.*, Los Angeles County Hospital).

Tipton (University of Tennessee, Knoxville) described a statistical analysis of trace metals in human tissues with particular emphasis on the lung. The report was based on findings from nine cities in tissues from 173 persons who were victims of instantaneous death. Twenty-nine tissues were analyzed by emission spectrography for 24 elements. Statistical analysis of the data used nonparametric methods since values were not distributed normally. Correlation patterns differed according to whether elements were "essential" or "nonessential."

Rustagi (University of Cincinnati) discussed the stochastic behavior of trace substances as they are taken up and excreted by the body. The distribution of most pollutants and their urine and serum levels are log-normal rather than normally distributed. The queueing theory should be applicable to these processes.

Bacaner (University of Minnesota) noted that several physiologic mechanisms are involved in the distribution of absorbed pollutants. He emphasized

the effects of the chemical nature of the pollutant, the size and physical state of the material, the portal of entry, chemical and physical interaction within the body tissue, utilization, damage or interference with normal mechanisms, and excretion mechanisms. Numerous illustrative examples were given.

Data on the body burden of long-lived isotopes are used to derive permissible dosages to body organs of persons occupationally exposed to ionizing radiation (Morgan, Oak Ridge National Laboratory). Accurate measurements of the body burden at postmortem examination are important because, currently, many assumptions have to be made due to the lack of data available on which to base estimates and calculations. Formulas relating body burden with intake and output were presented and the rationale for using "relative biological effectiveness" and "quality factor" was discussed.

Nelson (Institute of Industrial Medicine, New York University) noted carcinogenic implications of inhaled pollutants with particular emphasis on cancer of the lung. Intrinsic cellular sensitivity to the carcinogen and factors which modify effective dosage to target cells were indicated as important parameters. The relationship between the accumulated tissue burden of the carcinogens, chromate ion, and polynuclear hydrocarbons was discussed. Chromate ion appears to remain in the lung even though no correlation is shown between the lung burden of chromium and the occurrence of lung cancer; however, polynuclear hydrocarbons are rapidly cleared from the lung. Of even greater transiency is roentgen or gamma radiation. Some evidence suggests that proximal carcinogens react with nucleic acid and produce subtle modifications of DNA structures. The importance of particle size and the influence of structural alterations of the lung were stressed. Experimental bronchogenic cancer from the decay of ruthenium-106 occurred regularly and reproducibly with latent periods; it varied inversely with dosage and in frequencies increasing with dosage.

Gofman (Donner Laboratory, Division of Medical Physics and Lawrence Radiation Laboratory, University of California, Berkeley) described x-ray spectrochemical methods for studying the concentrations in serum of the elements between atomic numbers 15 and 92. Elements such as phosphorous, sulfur, chlorine, potassium, and calcium had high concentrations of from 100 ppm to 5000 ppm. Bromine, iron, copper, and zinc had serum levels of 1 ppm to 5 ppm, and virtually all the other elements of an atomic number greater than 15 showed serum levels well under 1 ppm in essentially healthy persons.

Dixon (Antioch College; chairman, Air Conservation Commission, AAAS) presented a special lecture on public policy in air conservation. He said that, "unregulated use of the atmosphere for waste disposal is an affront to human conditions; it tends to lower man's biological efficiency; it offends the senses; it damages his crops; it may adversely affect his longevity and ultimately may affect the processes of the evolution of the species." He stated it would be theoretically possible to build a city with no air pollution. However, on a more practical plane, it is also possible to regulate the pollutant levels in the air, but as is so often the case, public policy falls short of its responsibilities. In order to accelerate acceptance of necessary controls, Dixon mentioned the importance of scientists becoming involved in the discussions of public policy. Legislators should also remain receptive to radical innovations such as the nationalization of air space, new systems of mass transportation, and artificial ventilation of cities. He concluded by saying that the cost of clean air should be incorporated in our economic system and that clean air is not something which automatically can be taken for granted.

The session on laboratory and epidemiological relationships of respiratory disease and pollutant exposures was opened by Boren (Veterans Administration Hospital, Houston) who spoke on carbon as a carrier mechanism for irritant gases. Working with Swiss albino mice, activated carbon, and nitrogen dioxide, he showed that control mice and mice exposed to carbon alone exhibited essentially no structural changes and that survivors from exposures to nitrogen dioxide did not show any changes after an interval of 5 to 7 days. But animals exposed to carbon which had been permitted to absorb the nitrogen dioxide showed focal changes; this was apparently the result of tissue destruction at the alveolar level.

Results of work on continuous low-level exposures of rats to nitrogen dioxide were presented by Freeman and Haydon (Stanford Research Institute, Menlo Park, California). Rats exposed continuously to 50, 25, and 12 ppm developed voluminous, heavy lungs, and impairment of weight gain. The lungs showed increased glandular secretion and accumulation of PAS-positive material on the surface of the bronchial epithelium. The possible relationship of these changes to emphysema was discussed.

Balchum (University of Southern California School of Medicine) reported on immunologic mechanisms in emphysema. Severe interstitial pneumonitis developed in guinea pigs injected with lung tissue homogenate from other guinea pigs that had been exposed to nitrogen dioxide and nitric oxide. Positive agglutinations against lung tissue antigen developed. Similar reactions to latex particles coated with extracts of human lung tissue occurred in patients with lung cancer, tuberculosis, and emphysema; this did not happen in normal control subjects.

In reporting on immunologic methods in air-pollution research, Josephson (Laboratory of Medical and Biological Sciences, Division of Air Pollution, Public Health Service, Cincinnati) noted that skin sensitivity had been produced in animals after injection of micro-molar amounts of nitro-olefins mixed with Freund's adjuvant. Preliminary investigations of ozonated proteins and studies of protein fractions in tears were also discussed.

Two speakers, Toyama and McKerrow, discussed aspects of air pollution in their own countries, Japan and Great Britain, respectively. Toyama (Department of Preventive Medicine and Public Health, Keio University, Tokyo) reviewed the relationship of air pollution and health. Sampling and analytical instrumentation are not as well advanced in Japan as in this country, but sampling has been carried out using lead peroxide candles, dustfall gauges, and a Hemeon-type of tape sampler. The population growth in Tokyo has been associated with an increase in severe smog. The mortality rates from respiratory disease are greater in areas where there are many factories; morbidity surveys reveal similar findings. Pulmonary fibrosis on autopsy is found more frequently in persons residing in areas of heavy pollution. Impaired respiratory function was studied in school children who were examined repeatedly. When comparisons were made in children

residing in polluted and nonpolluted areas, the children residing in polluted areas had lower lung function during periods of heavy pollution. Experimental inhalation of dust deposited in polluted areas of Kawasaki City led to increased airway resistance in healthy humans. This is reversed by bronchodilator drugs. While classical bronchial asthma occurs predominantly in the autumn, about the same time of year that so-called "Tokyo-Yokohama Asthma" occurs, the highest number of bronchial disorders of children occurs in the spring at the same time of the highest levels of air pollution.

McKerrow (Pneumoconiosis Research Unit, Llandough Hospital, Wales) investigated the causes of chronic respiratory disease. Epidemiological studies have revealed an increase in morbidity due to chronic bronchitis in urban populations as compared with rural populations; this increase is accompanied by a lower ventilatory capacity. The prevalence of persistent cough and sputum was related to the number of cigarettes smoked. In men aged 55 to 64 a reduction in mean ventilatory capacity as related to the number of cigarettes smoked was also demonstrated. In laboratory studies, normal subjects exposed to coal dust for periods of 4 hours, 19 mg per cubic meter of air, showed an increase in respiratory resistance, but values had returned over half way to normal 1 hour after exposure. A 3-year prospective study of the ventilatory capacity in a group of ex-miners with pneumoconiosis and chronic bronchitis showed an annual cyclical change; the lowest and highest values were found early in February and August, respectively.

Studies of respiratory diseases and air pollution in Los Angeles were made by Oelsner, Wehrle *et al.* (Infectious Diseases Laboratory, University of Southern California). The prevalence of asthma was compared with cases in New Orleans in order to detect similarities, however none existed. School absenteeism and symptoms and signs of acute respiratory illnesses were studied in various groups. Work is still under way and a large number of relevant variables have been examined. No clearcut relationship to air pollution has been observed as yet.

Weill *et al.* (Tulane University School of Medicine) and Horton and McCaldin (Public Health Service, Division of Air Pollution) collaborated on further studies of asthma in New Orleans. Previous outbreaks of asthma have been related to point sources in city dumps in which underground combustion occurs. An increase in gaseous atmospheric pollution occurred on 5 October 1962, followed by an outbreak of asthma between the 7th and 10th of October. Extracts of the smoke from the burning dumps were analyzed by intradermal and scratch tests both on persons who had asthma during the outbreak and on students. The students who had a history of asthma or hay fever had more positive skin reactions than those without. Subjects who had current symptoms had a higher percentage of positive reactions than those whose symptoms were not active, and the persons who had episodes of asthma in the outbreak had a larger proportion of reactions than among the students.

The papers and discussion at the conference will be published in the near future in the *Archives of Environmental Health.*

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Electron Microscopy: Proteins in Experimental Pathology

Among the many sessions of the 47th annual meeting of the Federation of American Societies for Experimental Biology in Atlantic City (18 April) was a symposium on Electron Microscopy of Proteins in Experimental Pathology which was sponsored jointly by the Intersociety Committee for Research Potential in Pathology, Inc., and the American Society for Experimental Pathology.

In discussions on electron microscopy of the fibrinogen molecule and the fibrin clot, C. E. Hall (Massachusetts Institute of Technology) focused attention on changes that take place as fibrinogen molecules polymerize. Some years ago, Hall and Slayter proposed a model for fibrinogen, which consisted essentially of 3 beads, about 65 Å in diameter, that were held together by very thin strands; the length of the molecule was about 230 Å. More recent electron micrographs have indicated that at *p*H well above the isoelectric point, the average molecule of bovine fibrinogen appears to increase in length. A minimum length of approximately 230 Å was observed at the isoelectric point (*p*H 5.6) while at *p*H 11.9 the length averaged about 400 Å. The proposed model appears to be consistent with molecular weight data obtained from other sources. In a brief consideration of intermediate polymers of fibrinogen, Hall pointed out that polymerization appears to be end-to-end. Electron micrographs of fibrin filaments in clots have revealed that, in addition to the axial period of about 230 Å, there are intermediate bands. The fine structure of soluble fibrin closely resembles that of polymerizing fibrinogen, that is, there are units indistinguishable from fibrinogen molecules as well as end-to-end aggregates like those of intermediate fibrin polymers. While recent x-ray diffraction work by other researchers has confirmed the presence of the major periodicities in fibrinogen and fibrin as observed by electron microscopy, the relative intensities of diffraction orders cannot as yet be explained on the basis of electron microscopy; the x-ray findings may indicate the presence of substructure in the units observed by Hall.

S. J. Singer (Univ. of California, San Diego) discussed the development and present status of ferritin-antibody conjugates as stains for electron microscopy. The method, conceived and first developed by him and his co-workers, depends upon the chemical coupling of the ferritin (highly electron-scattering) molecule with antibody. At first, Singer used *m*-xylene diisocyanate as coupling reagent, and showed that such conjugates were only partly linked by covalent bonds. Use of another coupling reagent, toluene 2,4-diisocyanate, results in ferritin-antibody conjugates that are linked only covalently. Methods and controls necessary to check activities and specificities of ferritin-antibody conjugates were presented, and sources of error that have often been overlooked were pointed out. When an antigen to be reacted with conjugated antibody is in solution or in colloidal dispersion, the localization of the tagged antibodies can be quite specific. This is evident from electron microscopy of, for example, particles of tobacco mosaic virus or of influenza virus after contact with specific, conjugated antibodies. In spite of encouraging and significant results obtained in a number of laboratories, the application of the technique to the localization of material in sectioned cells is still encumbered by many unresolved problems—non-specific adsorption of specific conjugates, or of