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A System for Exposure of Mice to an Atmosphere Containing Carbon Particles

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A simple laboratory system and method of exposing small animals to varying concentrations of carbon particles in the atmosphere are described. The system requires no expensive, special equipment and, at the same time, permits selection of different particle-size ranges and particulate concentrations. Carbon particles may function as carriers of gaseous toxicants and thus the system permits of studies of this mechanism.

Introduction

INTEREST in air pollution and relation of pollutants to chronic lung disease stimulated our investigation of the interaction of gaseous and particulate material and the relation of this interaction to lung injury. Activated carbon was chosen as the particulate material because: (a) it is an active absorbing agent, (b) it is easily identified in tissues, (c) it is available in particle sizes which penetrate to the alveolar level,[1] and (d) it resembles the pigment frequently present in pulmonary emphysema.[2]

The present paper describes a system for the exposure of mice to an atmosphere containing carbon particles.

Materials and Methods

Activated carbon used for animal exposure is heated at 130°C for four hours and then stored over a desiccant until used. The resulting dry preparation is easily suspended in air.

Air is delivered into the system from a compressed air tank through a pressure regulator. The air passes through a flow meter and a desiccator, and then enters the carbon mixing chamber (Figure 1). The carbon mixing chamber is a five-liter, round-bottom flask containing the rotor of a magnetic stirrer and the dry carbon preparation. The dry air entering the chamber through a capillary tube is directed into the agitated carbon and a suspension of carbon in air is formed. The suspended carbon is swept into the animal exposure chamber by the air leaving the carbon mixing chamber. A minimum air flow of two to three liters per minute through the mixing chamber is required to maintain the carbon suspension. A metered supplementary air flow enters the system between the carbon mixing chamber and the animal exposure chamber allowing further control over carbon concentration by varying the amount of dilution with clean air. The supplementary air enters the main line through a glass tip which projects into the main air flow and curves toward the animal exposure chamber.

The animal exposure chamber is constructed from a 250-mm desiccator with a 55/48 tubulation in the lid. As detailed in the inset (Figure 1), the aspirator sleeve is replaced with a glass head through which pass two tubes. The carbon suspension enters through the short tube and is released into the upper part of the exposure chamber. A longer tube extends beneath the supporting screen for the animals and is the exit for air passing through the chamber. The exposure chamber contains water and food for the animals. Carbon in the exhaust from the animal chamber is removed by a water trap before the air is released into a ventilated hood.

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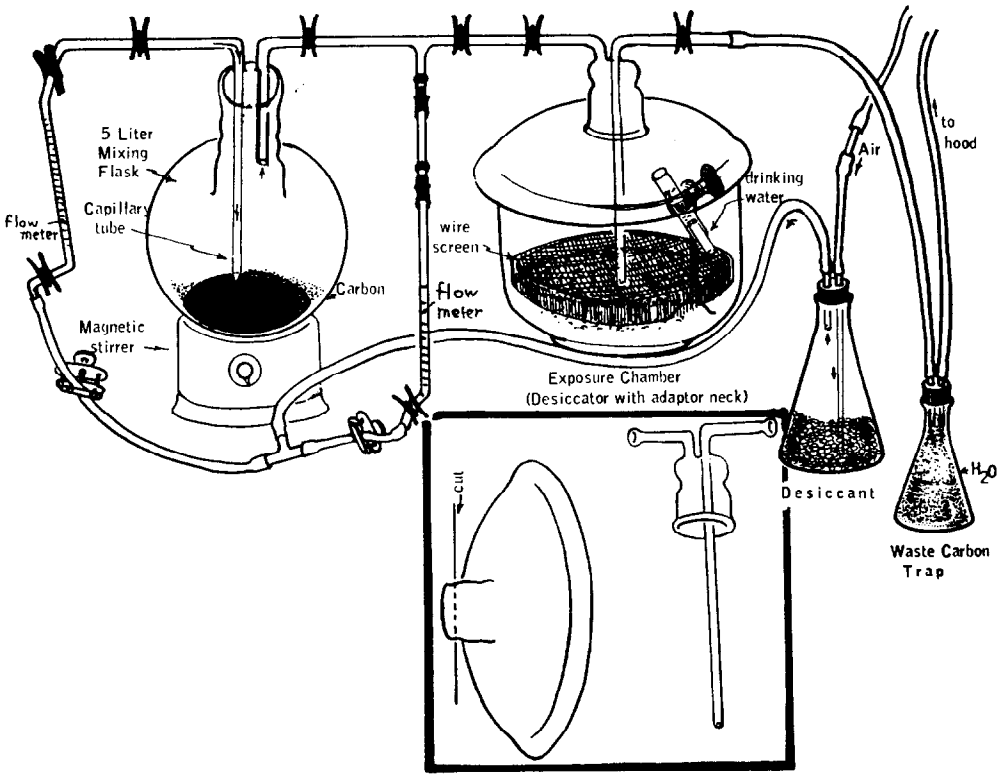

FIGURE 1. Sketch of apparatus for the dispersal of carbon particles in air and exposure of small animals.

In operation, a maximum of twenty mice is placed in the exposure chamber and the top is secured. After the magnetic stirrer is started, the air flow through the carbon mixing chamber is adjusted to give the desired carbon suspension. Further control of carbon concentration is accomplished by varying the amount of supplementary air. Visual estimation of density of carbon suspension is satisfactory for qualitative study.

After exposure the mice are held in the normal animal colony for observation until sacrificed for histologic study. The lungs are fixed in expansion by the method of fume fixation.[3] The study of thick sections embedded in plastic with or without counter stain illustrates the distribution of carbon particles.

Discussion

The system described is moderately selective of the range of particle sizes which are carried into the exposure chamber. The mean diameter of 100 particles prior to suspension as measured with an ocular micrometer was 16.3 microns with a standard deviation of 14.3 microns. The mean diameter of 100 particles recovered as they entered the animal exposure chamber was 10.6 microns with a standard deviation of 6.5 microns.

A total number of 53 mice have been exposed to suspensions of carbon in air for six hour periods with a total time of exposure ranging from 6 to 640 hours. There has been an occasional unexplained death in the exposure chamber. There has been no evident morbidity during a post exposure survival period of one year.

Examination of lungs of mice surviving exposure for twenty-four hours to nine months shows distribution of carbon throughout the lung at the alveolar level. With increasing time of survival after exposure there is increased localization of carbon pigment in the

regional peribronchial nodes. No evident lesion has been associated with the deposition of the pigment in the lung.

The distribution of carbon pigment at the alveolar level and the innocuous nature of this substance suggest that it is a satisfactory carrier for use in the investigation of the effect of absorbed or adsorbed toxicants.

Summary

A system for the exposure of mice to suspensions of carbon particles in air is described and illustrated. Mice have tolerated heavy suspensions of carbon for long periods with no evident morbidity during a period of one year following exposure. It is suggested that such carbon particles might function effectively as carriers for gaseous toxicants.

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