The experimental data also suggest that lung lesions may be present long before they are clinically apparent. Classically, pulmonary complications become apparent only about a week after paraquat ingestion, but lesions are regularly apparent in animals long before this.

At present, it is still not clear from our data whether the airway epithelium is involved or not and we are studying the matter further. As noted, rather nonspecific alterations were seen in some paraquat animals, but not in all of them, and it is clear that airway epithelium shows lesions which, if real, are far less obvious than those seen in alveolar epithelium. Absence of lesions in airway epithelium would be of considerable interest. It is not clear why certain organs and cells are particularly sensitive to paraquat. The similarity to oxygen poisoning and the additive effect of oxygen to paraquat poisoning has suggested that paraquat may particularly affect the lung because of its high ambient oxygen. Were this the case, then airway epithelium should be especially involved but this is not the case. Alternatively, cells may be particularly sensitive because paraquat goes selectively to them. This is a hard question to solve in the lung interstitium where the epithelium appears sensitive and the endothelium relatively resistant. Paraquat is highly soluble in water and thus ordinary autoradiographic techniques cannot be used and, in addition, high resolution is necessary to separate endothelium from epithelium. However, the presence or absence of radioactive paraquat in airway epithelium should be relatively easy to determine and we are at present examining this problem.

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Pathophysiology of Experimental Canine Interstitial Lung Disease*

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Patients with diffuse interstitial lung disease, characterized by restriction of lung volumes, impaired diffusion and decreased compliance, frequently manifest ventilatory disturbances such as dyspnea, tachypnea and alveolar hyperventilation. In the milder stages of the diseases, these abnormalities of ventilation may be present only during exercise. The physiologic mechanism underlying such excessive ventilatory drive has not been identified, but could involve neural stimuli arising in bronchopulmonary sensory receptors whose afferent pathways are in the vagus nerves. Previous attempts to investigate this possibility have generally involved studies on anesthetized animals in whom acute bronchopulmonary disturbances have been produced, such as the imposition of an external respiratory elastic load applied to the airway. There are both practical limitations and theoretical objections to this approach. Accordingly, we have examined the respiratory physiologic disturbances that occur in conscious dogs during the course of interstitial lung disease induced by the intravenous administration of complete Freund's adjuvant.

Material and Methods

Our studies were performed on four dogs trained to stand quietly and to run on a treadmill. Once trained they underwent surgical preparation that consisted of creation of an exteriorized cervical vagal loop on each side of the neck. The carotid arteries were placed in a subcutaneous position to make them readily accessible for future cannulations prior to each study. Finally, a permanent tracheostomy was created to allow the later insertion of an endotracheal tube during the studies. Following surgery, at least one month was allowed for healing. All studies were performed with the dogs conscious. Initially, the dogs were studied repeatedly in the healthy state over a three-month period to establish normal values of each of the variables for each dog, since each dog served as his own control for the disease studies.

Results

Following the control studies, interstitial lung disease was induced in the dogs by the intravenous administration of complete Freund's adjuvant, 0.3 ml/kg on each of two days. The physiologic measurements were repeated at frequent intervals throughout the course of the disease, a period of six to eight weeks, following which there was complete recovery. Histologic studies were performed on eight other dogs whose lungs were removed at varying intervals following injection of Freund's adjuvant. The pulmonary reaction to the adjuvant consisted of two temporally distinct phases. One week following injection there was a diffuse interstitial pneumonitis characterized by edema and increased cellularity of alveolar walls. The cells consisted predominantly of histiocytes, but many lymphocytes, plasma cells and polymorphonuclear leukocytes were also present. The histiocytes stained heavily for lipid which was presumably derived from the paraffin oil of the Freund's adjuvant. However, no lipid-staining material could be detected in the pulmonary blood vessels. By two weeks following injection, the histologic picture had changed considerably. The pulmonary abnormality now was a proliferative granulomatosis, resembling sarcoido-
sis, as described by Strauss, Caldwell and Fritts. The predominant cell in the granulomas was a large mono-nuclear, epithelioid cell with a foamy, vacuolated cytoplasm. These cells stained heavily for lipid. Similar granulomas were found in the hilar lymph nodes, spleen and liver. The granulomatous phase of the disease persisted for a further two to three weeks and thereafter regressed.

The physiologic measurements at rest included total lung capacity (TLC), functional residual capacity (FRC) and diffusing capacity for carbon monoxide (DLco). DLco was determined by the single-breath technique and lung volumes by a neon dilution method. The ten-second breath hold required for the DLco measurement could be obtained in conscious dogs since maintained inflation of the lungs results in apnea, the Hering-Breuer inflation reflex. During both phases of the interstitial lung disease, there were significant decreases in TLC, FRC, and DLco, but a return to the dogs' normal values by six weeks following injection of the Freund's adjuvant.

In addition to measurements of lung volumes and diffusion, we obtained static deflation pressure-volume curves of the lungs by training the dogs to swallow an esophageal balloon-catheter. Static conditions were achieved by again taking advantage of the inflation reflex. At two weeks following injection there was a marked shift to the right and decrease in slope of the pressure-volume curve, and over the subsequent four weeks there was a gradual return towards normal so that by six weeks the values fell within the normal range.

Ventilatory measurements were made at rest and during steady-state exercise (treadmill running). During both phases of the interstitial lung disease, the minute volume of ventilation (Vt) was normal at rest, but increased significantly beyond the normal range during mild exercise. Respiratory frequency (f) was abnormally elevated at all levels of work and tidal volumes (Vt) were significantly decreased. One dog was hypoxic at rest, as compared to the healthy state, but the other three dogs were not. During mild exercise, the hypoxemia became more severe. Alveolar ventilation, as indicated by the arterial CO2 pressure, was normal at rest, but with mild exercise all the dogs hyperventilated and lowered the CO2 pressure dramatically, resulting in a respiratory alkalosis.

Thus, all of these various results indicate that the physiologic disturbances in these experimental interstitial lung diseases in dogs resemble those found in many pulmonary parenchymal diseases in man. Accordingly we next investigated the origin of the exercise hyperventilation by repeating the studies with both vagus nerves blocked completely. Vagal blockade was accomplished by cooling the vagal loops with external radiators, as described previously. In every case there was a significant reduction in Vt during exercise down into the normal range for each dog, indicating that vagal impulses had been responsible for the abnormally high Vt when the vagi were intact. This effect of vagal blockade in the diseased dogs was in marked contrast to the healthy dogs in whom vagal blockade produced no decrease in Vt. Furthermore, vagal blockade slowed f markedly and increased Vt, indicating that the abnormally rapid and shallow breathing during disease was significantly dependent upon vagal impulses. However, even with the vagi blocked, breathing remained more rapid and shallow than when the vagi were blocked during health, suggesting that extra-vagal mechanisms were also contributing to the abnormality of ventilatory pattern.

In contrast to these effects of complete vagal blockade, that is sensory and motor, we examined the effects of blocking only the vagal motor nerve endings with atropine sulphate, 0.15 mg/kg, iv. Despite adequate evidence of complete motor blockade, there were no significant changes in the volume or pattern of breathing at rest or during exercise. Thus, the profound changes observed during vagal cooling must have been due to blockade of vagal sensory fibers.

Finally, we examined the possible role of hypoxemia in contributing to the exercise hyperventilation by repeating the studies while the dogs breathed sufficient O2 to raise the arterial oxygen pressure to 300 mm Hg. Whether the vagi were intact or blocked, breathing oxygen produced no significant decrease in Vt and no change in f or Vt. Thus, hypoxemia did not appear to be a major drive to the excessive ventilation.

We have demonstrated in conscious dogs that the physiologic disturbances in diffuse interstitial pneumonitis and in proliferative pulmonary granulomatosis simulate those seen in many patients with interstitial lung disease. These disturbances include restricted lung volumes, impaired diffusion, decreased compliance, and exercise hyperventilation with rapid shallow breathing. Furthermore, we have shown that sensory impulses mediated by the vagus nerves account to a major extent for the excessive ventilatory drive that exists in these disease models. These findings suggest that vagal impulses could account for the dyspnea, tachypnea and hyperventilation characteristic of interstitial lung disease in man.

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