The Blood-Air Barrier in Pulmonary Asbestosis: Study of a Case by Electron Microscopy


A patient is described in whom pulmonary asbestosis was proved by open-lung biopsy. In the specimens examined, extensive inflammatory and fibrotic changes were present and ferrugineous bodies were demonstrated by iron stains. Characteristic reduction in lung volumes and diffusing capacity were improved by avoidance of exposure to asbestos, most probably because of reversibility of inflammatory changes in alveoli. Within the limits of the sampling method, electron microscopy showed that the disease involved principally alveolar septal interstitium. The appearance of the surviving blood-air barriers was normal.

Thickening of the alveolar-capillary membrane has been considered a major cause of the physiologic alterations in interstitial pulmonary fibrosis, and patients with this condition have the signs, symptoms, and abnormalities of pulmonary function characteristic of the "alveolar-capillary block" syndrome. Asbestosis, which has been increasing in frequency during the past 70 years, is one of the causes of pulmonary fibrosis of this type. Previously, we have reported that examination of lung tissue from patients with diffuse pulmonary fibrosis demonstrated that morphologically normal alveolar-capillary membranes were present in the samples studied. These structures could not be examined in detail before the advent of electron microscopy. The present study was undertaken to examine the fine structural appearance of the blood-air barriers in a sample of lung tissue from a patient with pulmonary asbestosis.

CASE REPORT

A 26-year-old Caucasian asbestos worker came to the Mayo Clinic in February 1964. His primary complaints were shortness of breath and chest pains of three to four months' duration. He had been in excellent health until October 1963, when he noted dyspnea on exertion. Subternal pressure also had been present with exertion, but he had been free of chest pain for a month before his initial visit, although exertional dyspnea persisted. A chronic cough productive of one tablespoonful of yellowish sputum each day had troubled him for the previous year, and on many occasions, the sputum had been streaked with blood. There was no wheezing, dependent edema, hypertension, orthopnea, or paroxysmal nocturnal dyspnea. His weight had been stable and his appetite good. There were no symptoms other than the pulmonary complaints. For five years before the pulmonary symptoms began, he had been employed as a "lagger" or pipe wrapper in the asbestos industry. Fiberglass also was used. However, the patient never had taken the precautions recommended for the prevention of inhalation of these agents. He had smoked 30 cigarettes daily for most of his life. There was nothing of significance in his previous medical history or family history.

Significant physical findings were limited to the chest. The posterior lung bases were dull to percussion, and breath sounds were diminished in these areas. On deep breathing, a pleural friction rub was heard over the upper left anterior thorax.

Blood hemoglobin level, total and differential leukocyte counts, serologic findings, lupus erythematosus clot test, fasting blood sugar, blood urea, urine, and serum protein electrophoresis were all normal or negative. An electrocardiogram was within normal limits. Intermediate and second-strength tuberculin skin tests were negative. A histoplasmin skin test was negative in a dilution of one in ten.

Blunting of both costophrenic angles was noted on the chest roentgenogram, and decubitus views revealed that this was due to pleural thickening. There was partial collapse of the right middle lobe, and small areas of fibrosis or pneumonitis were apparent in the superior and posterior basal segments of the left lower lobe (Fig 1). In three sputum smears,
no acid-fast bacilli or fungi could be found, and results of subsequent cultures were negative for these organisms.

Lung volumes and spirometric tracings were obtained with a spirometer of the Benedict-Roth type, and residual volume was determined by a modified nitrogen-elimination technique. Normal values for lung volumes were estimated from Bateman's formula, and for maximal breathing capacity from the Baldwin formula. Estimated normal values for maximal midexpiratory flow were derived from the data of Leuallen and Fowler. Arterial blood-oxygen saturation was determined by the use of an earpiece oximeter at rest and during exercise while breathing room air and oxygen. Carbon monoxide-diffusing capacity studies were undertaken with the use of a steady-state method, and values were obtained during exercise.

Small lung volumes without significant reduction in airway dynamics were consistent with a predominantly restrictive pattern of lung disease. Steady-state carbon monoxide-diffusing capacity was reduced below normal limits during exercise, indicating an abnormality of blood-air gas transfer. Arterial blood oxygen saturation was normal at rest and did not desaturate on exercise, when measured by earlobe oximetry (Table 1).

Because the diagnosis was not definite, a bronchoscopic examination was carried out. All major bronchial segments were well visualized and appeared to be normal. No source of bleeding could be found. There were no mycobacteria or fungi in the smears or in subsequent cultures of bronchial washings obtained at this time from which only streptococci of the viridans type was grown. Subsequently, an open lung biopsy was performed, and tissue from the right middle lobe and pleura was obtained. Specimens of tissue were processed and examined by light microscopy and by electron microscopy, and other portions were submitted for cultures for mycobacteria, fungi, and aerobic and anaerobic bacteria; all results subsequently were negative. Results of examination by light microscopy were consistent with a diagnosis of pulmonary asbestosis.

Four months later, the patient returned for evaluation. In the interval, he had done well and had rarely worked with asbestos. He continued to have a mild unproductive cough but had no further episodes of hemoptysis. His exertional dyspnea had not changed. His examination was not remarkable except for a few inspiratory rhonchi over the right midlung field which cleared with coughing. A chest roentgenogram appeared little changed. Pulmonary function data at his second admission showed improvement (Table 1).

<table>
<thead>
<tr>
<th>Measurement</th>
<th>Estimated Normal</th>
<th>February 1964</th>
<th>July 1964</th>
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</thead>
<tbody>
<tr>
<td>Static lung volumes (liters)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Vital capacity</td>
<td>5.2</td>
<td>3.2 (3.2*)</td>
<td>4.18 (4.04*)</td>
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<tr>
<td>Residual volume</td>
<td>1.8</td>
<td>1.95</td>
<td>1.57</td>
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<tr>
<td>Total capacity</td>
<td>7.0</td>
<td>5.16</td>
<td>5.75</td>
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<tr>
<td>Residual volume/total capacity × 100</td>
<td>&lt;30</td>
<td>38</td>
<td>27</td>
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<tr>
<td>Functional residual capacity</td>
<td>3.2</td>
<td>3.52</td>
<td>3.62</td>
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<tr>
<td>Expiratory reserve</td>
<td>1.4</td>
<td>1.57</td>
<td>2.05</td>
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<tr>
<td>Maximal breathing capacity (liters/minute)</td>
<td>140</td>
<td>161 (134*)</td>
<td>118 (126*)</td>
</tr>
<tr>
<td>Maximal midexpiratory flow (liters/second)</td>
<td>&gt;2.5</td>
<td>2.0 (2.5*)</td>
<td>2.5 (3.3*)</td>
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<td>Nitrogen washout index (%)</td>
<td>≤2.5</td>
<td>0.6</td>
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<td>Arterial oxygen saturation (%) †</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Resting (air)</td>
<td>95-98</td>
<td>96</td>
<td>96</td>
</tr>
<tr>
<td>Resting (oxygen)</td>
<td>100</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>Before exercise (air)</td>
<td>95-98</td>
<td>97</td>
<td>96</td>
</tr>
<tr>
<td>Exercise (air)</td>
<td>No decrease</td>
<td>96</td>
<td>97</td>
</tr>
<tr>
<td>Exercise (oxygen)</td>
<td>100</td>
<td>100</td>
<td>100</td>
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<tr>
<td>Ventilation</td>
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<tr>
<td>Liters/minute</td>
<td></td>
<td>9.81</td>
<td>10.42</td>
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<tr>
<td>Rate (respirations/minute)</td>
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<td>15.0</td>
<td>15.6</td>
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<tr>
<td>Tidal volume (ml)</td>
<td></td>
<td>654</td>
<td>668</td>
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<tr>
<td>Carbon monoxide-diffusing capacity at steady-state, exercise (ml/min/mm Hg)</td>
<td>32 (24-40)</td>
<td>23</td>
<td>26</td>
</tr>
</tbody>
</table>

*After 1 ml nebulized isoproterenol, 1:400.
†Measured by ear oximeters.
Histopathologic Features

Lung tissue for light microscopy was fixed in 10 percent neutral formalin, and sections were prepared by standard methods. Staining was carried out with hematoxylin and eosin, and with elastic tissue-van Gieson, Mallory-Heidenhain, and iron stains.

For electron microscopy, fixation was carried out promptly with Dalton's solution at a pH of 7.6, and the tissue was embedded in an epoxy resin (Epon 812) by the method of Luft. Sections were cut with a diamond knife, mounted on copper grids, and stained with uranyl acetate.

Light microscopy of the pleura showed mild inflammatory changes, with minimal fibrosis and focal areas of calcium deposition. The lung parenchyma revealed chronic interstitial pneumonitis and peribronchiolar fibrosis with focal intra-alveolar collections of pigment-laden macrophages and lipid material. Scattered asbestos bodies were demonstrated by hematoxylin and eosin and by iron stains (Fig. 2).

At the higher magnifications of electron microscopy, the areas of severe interstitial pneumonitis and fibrosis seen under the light microscope were detailed further (Fig 3). Distortion of alveolar septa and infiltration by macrophages, fibroblasts, and lymphocytes were prominent features. Where septal damage was most severe, replacement by formed collagen and obliteration of septal capillaries occurred. In some of these areas, blood vessels were totally absent. Where capillaries remained, they were displaced to the periphery of the septum by cellular infiltration or collagen. There was no concentric pericapillary thickening, and morphologically normal alveolar-capillary membranes were found adjacent to alveolar spaces. These membranes varied from 391 to 2,154 μ in width and were, therefore, in the normal human range of 390 to 2,078 μ previously described by ourselves and others.11-13

Comment

Experimental studies have shown that tissue reaction in the lung of the guinea pig can be detected as early as two weeks after exposure to asbestos dust. The earliest change is an inflammatory response in terminal bronchioles and adjacent alveoli.14 Inflammatory cells and exudate can be seen in the lumen of involved bronchioles and nodular giant cell lesions in their walls. Increased numbers of alveolar macrophages appear, and coarse fibers of asbestos may be visible in their cytoplasm or be free in tissue spaces. Special stains may demonstrate formed collagen and an envelope of iron-containing protein around some of the dust particles. With the appearance of collagen, the focal interstitial pneumonitis originally present in relation to the bronchiolar lesions is replaced by a confluent interstitial fibrosis. Adenoid proliferation of bronchiolar epithelium develops, with polypoid outgrowth into the lumen. Alveolar walls are greatly

Figure 2. A, Interstitial pneumonitis with macrophage in center of field, containing single thin asbestos fiber (hematoxylin and eosin; reduced from x 520). B, Lung tissue showing ferruginous bodies (iron; x 370).
thickened, and macrophages and giant cells are prominent. Typical asbestos or ferruginous bodies indicate a protective reaction to the fibrogenic particles of asbestos dust.\textsuperscript{15} Although this description of the changes seen in developing pulmonary asbestosis is based on experimental work, the disease in the human probably progresses in a similar fashion.

The immediate cause of these changes is not certain. Physical or chemical irritation, autoimmune disturbance, and release of fibrogenic substances by disintegrating phagocytes have been proposed, but none of these theories is completely accepted.\textsuperscript{16,17} For many years, it was believed that only large fibers of asbestos damaged the lung. However, electron microscopy has revealed recently that many more asbestos particles are involved in the pathologic process.\textsuperscript{14} In an electron microscopic study of guinea pig lung, Davis\textsuperscript{15} showed that the greater part of asbestos dust which penetrates the finer air passages has a very small particle size, much of it less than 1 μ long. The accumulated evidence indicates that smaller fibers are at least of equal importance in causing disease, and because of their greater penetrance, they may be more damaging to the finer air passages in which the initial changes occur.

The formation of asbestos or ferruginous bodies is also a significant protective factor in the development of pulmonary fibrosis from asbestos inhalation. There is convincing evidence available that as long as an asbestos fiber is encased in a protein and ferritin coating, it remains inactive and is not fibrogenic.\textsuperscript{18} Uncoated fibers are much more numerous and stimulate the production of collagen around them. However, ferruginous bodies are not numerous in areas of fibrosis and are best regarded as indicators of exposure to a precipitating dust. Their production occurs in the cytoplasm of a cell which does not demonstrate obvious alteration during this process.\textsuperscript{19}

The ultramicroscopic changes present in the diseased lung of our patient with clinical pulmonary asbestosis were similar to those we previously observed in idiopathic pulmonary fibrosis and pulmonary histiocytosis X.\textsuperscript{2,3} Alveolar walls were infiltrated with dust-laden macrophages, plasma cells, and eosinophilic leukocytes. In some areas, no recognizable alveolar structure could be seen, the normal architecture having been replaced by cellular infiltration or by collagen. Davis\textsuperscript{19,20} described changes in basement membranes in the blood-air barrier which we were unable to recognize in the present study. He also demonstrated, in experimental work, the ability of dust-laden alveolar macrophages to form giant cells by intertwining narrow, elongated, cytoplasmic processes to link several adjacent macrophages together. He concluded that these cells, either individually or as part of a giant cell, can become fibroblasts when the situation requires it.

Cellular infiltration or fibrotic replacement of alveolar walls reduced the number of alveolar septal capillaries seen. Those that remained were small and were displaced, by the tissue changes, to one side of the septum, where one wall was invariably adjacent to an alveolar space. As in idiopathic pulmonary fibrosis, the appearance and width of these alveolar-capillary membranes were normal. Although occlusion of capillaries by platelet thrombi has been described, we did not find a vessel in which this had occurred.

In patients with diffuse parenchymal lung disease, including asbestosis, the blood-air gas exchange is disturbed. Peripheral arterial oxygen saturation decreases during exercise and later in the course of the disease may be reduced at rest. Diminished lung volumes and compliance, absence of airway obstruction, hyperventilation at rest and during exercise, and a normal or low arterial carbon dioxide level are the accompanying physiologic derangements seen in these patients. Diffuse thickening of the alveolar-capillary membrane has been suggested as a major factor responsible for these changes.\textsuperscript{1} However, Finley and co-workers\textsuperscript{21} have calculated that failure of equilibration of oxygen in the alveolus is not essential in explaining the
peripheral arterial oxygen desaturation that occurs in these diseases because disturbances of the normal ventilation and perfusion relationships in the lung could produce the same result. The demonstration of increased physiologic dead space and nonuniform distribution of inspired gas has provided additional support for this concept.\textsuperscript{22}

A reduction in pulmonary diffusing capacity has been regarded as such a characteristic change in pulmonary asbestosis that surveys of asbestos workers are not regarded as complete unless this aspect of pulmonary function is measured.\textsuperscript{23} Diffusing capacity may be reduced before definite evidence of roentgenographic change has occurred, in keeping with the belief that the earliest lesion is an alveolar exudate.\textsuperscript{24} As the disease becomes worse, the abnormalities of diffusing capacity increase also. Ventilation-perfusion inequality exists in these patients, but the amount of the disturbance that can be ascribed to this inequality or to the decrease in lung volume or to qualitative changes in alveolar walls is still uncertain. Ventilatory capacity is well preserved unless compensatory emphysematous change develops. Reduction in pulmonary compliance and increase in the nonelastic work of breathing also have been demonstrated in these persons.\textsuperscript{25} In an extensive study, Hunt\textsuperscript{26} has shown that routine measurements of pulmonary function may demonstrate involvement of the lungs by asbestosis before evidence of roentgenographic change appears.

As is characteristic in asbestosis, the patient described in our present study had reduced total lung and vital capacities and a low steady-state carbon monoxide-diffusing capacity. We could not demonstrate diffuse thickening of the alveolar-capillary membranes and previously have reported finding normal alveolar-capillary membranes in samples of lung tissue from patients with diffuse pulmonary fibrosis who also had reduced carbon monoxide-diffusing capacities.\textsuperscript{23} The improvement in pulmonary function that occurred after exposure to asbestos was avoided is probably due to reversibility of early bronchiolitis and alveolitis.

Within the limits of the sampling method, our studies indicate that many normal blood-air barriers exist in this and other diffuse fibroses and that diffuse and uniformly distributed thickening of these barriers is not a frequent finding. In this type of study, it is not possible to quantitate reduction in the alveolar vascular bed or to determine whether the cells or fibrous tissue displacing or obliterating these vessels acts as an impediment to gas diffusion. Replacement of normal lung tissue by cells and fibrous tissue might result in regional changes in ventilation and perfusion of the lung, and no doubt this is important in the production of the physiologic abnormalities that occur. Methods are available for the quantitative measurement of lung structures and should be capable of resolving the problem of the distribution of such changes in lung parenchyma when used with light microscopy.

\textbf{REFERENCES}

18. Davis JMG: Electron-microscopic studies of asbestosis in
Predecessor of Sigmund Freud

Much work has been done in tracing the genealogy of the basic ideas that Freud employed in his psychology. It was a Polish psychologist, Louise von Karpinska, who first called attention to the resemblance between Freud's fundamental ideas and those promulgated by Herbart seventy years previously. Herbart's conception of the unconscious was the only dynamic one before Freud's. According to it, unconscious mental processes are dominated by a constant conflict which Herbart describes in terms of ideas of varying intensity—a notion which Freud later replaced by a conflict of affects. Herbart actually describes an idea as "verdrängt" (suppressed) when it is unable to reach consciousness because of some opposing idea or when it has been driven out of consciousness by one. "Science knows more than what is actually experienced (in consciousness) only because what is experienced is unthinkable without examining what is concealed. One must be able to recognize from what is experienced the traces of what is stirring and acting behind the curtains."

Jones E, in Olson R: Science as Metaphor—The Historical Role of Scientific Theories in Forming Western Culture, Belmont, California, Wadsworth, 1971