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Busulphan lung

W. A. LITTLER, J. M. KAY, P. S. HASLETON, AND DONALD HEATH

From the Cardio-thoracic Unit, Broadgreen Hospital, Liverpool, and the Department of Pathology, University of Liverpool

A 61-year-old man with chronic myeloid leukaemia was treated with busulphan (Myleran). After receiving 1 g. of this drug over a period of 20 months he became dyspnoeic and developed crepitations in the lungs. Two months later radiographs of the chest revealed peri-hilar infiltrates and subsequently diffuse mottling throughout both lungs. Lung function tests showed a gross impairment of the transfer factor to a quarter of the predicted normal. At necropsy the lungs showed a striking proliferation of granular pneumocytes, many of which had disintegrated to produce intra-alveolar debris, some of which showed organization by fibrous tissue. There was associated interstitial pulmonary fibrosis. Electron microscopy confirmed the desquamated alveolar cells to be type II (granular) pneumocytes containing characteristic lamellar bodies. Many of these osmiophilic bodies, believed to be the source of pulmonary surfactant, had been liberated into the alveolar spaces, with the formation of phospholipid myelin figures and lattices. We think that the basic pathology of busulphan lung is a chemically induced alveolitis with proliferation of granular pneumocytes followed by fibrosis of alveolar walls and intra-alveolar contents.

Here we describe in detail the clinical and pathological features of a patient who developed pulmonary complications following a course of busulphan (Myleran) for chronic myeloid leukaemia. Lung function studies were performed in addition to clinical examination. Electron microscopy of the lung was carried out as well as routine histopathology and has yielded new information on the pathology of 'busulphan lung'.

CASE REPORT

The patient, a 61-year-old male clerk, was first seen in January 1967 because of blurred vision in the left eye and tiredness for three weeks. On examination the mucous membranes and nail beds were pale, the spleen was enlarged five fingerbreadths below the left costal margin, and the liver was also palpable. There were bilateral retinal haemorrhages and exudates. The chest, heart, and nervous system were normal on clinical examination, and the systemic blood pressure was moderately raised at 180/105 mm. Hg.

The blood picture was typical of chronic myeloid leukaemia with a total white cell count of 256,000/cu. mm. A differential count showed blast cells 8%, myelocytes 36%, metamyelocytes 5%, neutrophils 46%, lymphocytes 2%, and eosinophils 3%. The platelet count was 129,000/cu. mm. and the haemoglobin level was 8·8 g./100 ml. (60%). Bone marrow examination was not carried out. Peripheral blood chromosome culture revealed the Philadelphia (Ph^1) chromosome. The chest radiograph was normal (Fig. 1).

A course of busulphan therapy was started on 13 January 1967. The initial dose of 4 mg. daily was reduced to a maintenance dose of 2 mg. four times a week on March 28, when he was in a satisfactory clinical and haematological remission with a total white cell count of 55,000/cu. mm., a platelet count of 465,000/cu. mm., and a haemoglobin level of 13·8 g./100 ml. (95%). This remission continued with doses of busulphan, 2 mg. four or five times a week, until 13 June 1968, when a total white cell count of 19,300/cu. mm. showed a differential count of blast cells 24%, myelocytes 7%, neutrophils 50%, lymphocytes 10%, monocytes 6%, eosinophils 1%, and basophils 2%. At this stage the platelet count was 150,000/cu. mm. and the haemoglobin level was 8·9 g./100 ml. (61%).

The patient's only complaint was tiredness and the main physical finding was splenomegaly. Because of the myeloblastic blood picture the dose of busulphan was kept at 2 mg. four times a week, and in addition a course of 6-mercaptopurine, 150 mg. daily, was started. On 9 July 1968 the haemoglobin level had fallen to 7·1 g./100 ml. (51%). He was transfused with four pints of blood and the dose of busulphan was tailed off and finally stopped on 26 July after 19 months' treatment with a total dose of

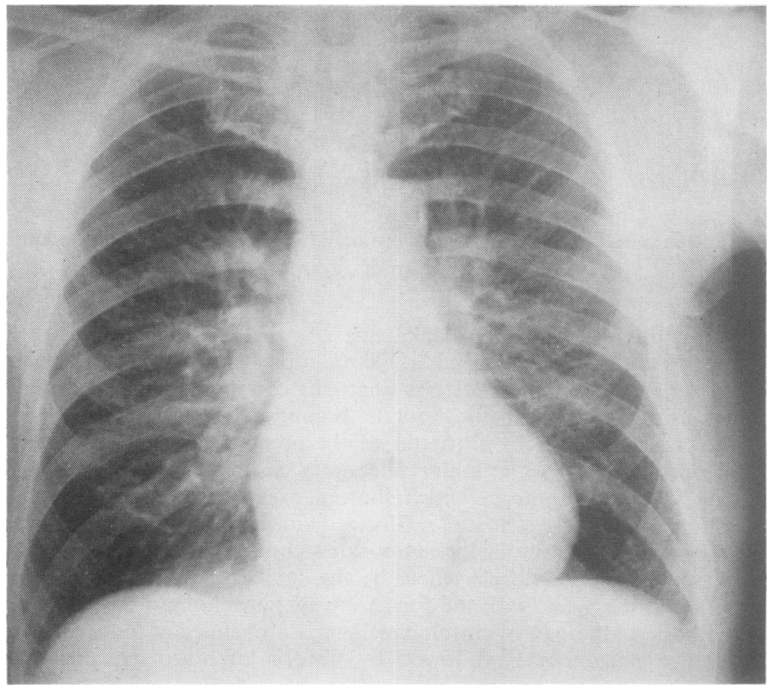

FIG. 1. *Normal chest radiograph taken in January* 1967 *when chronic myeloid leukaemia had been diagnosed.*

1,000 mg. A chest radiograph taken at this time was normal and unchanged.

In September 1968 the patient began to complain of dyspnoea on moderate activity such as climbing stairs or walking up an incline. On examination he had slight sacral and ankle oedema but no jugular venous engorgement. A soft apical pan-systolic murmur was heard, but the heart sounds were normal. Bilateral basal crepitations were present. These findings were attributed to cardiac failure associated with anaemia, since the haemoglobin level was 7·3 g./100 ml. (50%). The blood film showed anisocytosis and polychromasia with 9 normoblasts per 100 white cells. The reticulocyte count was 4%. The serum bilirubin was 1·1 mg./100 ml., but there was no excess of urobilinogen in the urine and Schumm's test was negative. The direct Coombs' test was negative. The chest radiograph and the electrocardiogram were normal. Four pints of blood were transfused and he was given 40 mg. frusemide each day. The patient's symptoms improved and the systemic oedema went, but the basal crepitations in the lung persisted. Since there was haematological evidence of haemolysis, prednisolone was added to the treatment in an initial dose of 40 mg. daily, reducing over a five-day period to 20 mg. daily.

During the next month his dyspnoea on effort increased rapidly, so that he became breathless even at rest and he also developed a non-productive cough. Examination on 4 October 1968 revealed tachypnoea, central cyanosis, bilateral basal crepitations, and a fever of 101° F. The blood film showed evidence of reduced myeloblastic activity, 8% of the total white cell count of 8,500/cu. mm. consisting of blast cells. The haemoglobin level was 10·8 g./100 ml. (74%). The patient was now considered to have a pulmonary infection and a week's course of ampicillin, 500 mg. six-hourly, was given. The temperature settled but the dyspnoea, central cyanosis, and basal crepitations persisted. On 6 November a chest radiograph showed peri-hilar infiltrates, particularly in the right sub-apical region, with an area of consolidation in the posterior segment of the left lower lobe (Fig. 2). He was treated with a transfusion of four pints of blood, tetracycline, 500 mg. six-hourly, digoxin, 0·25 mg. twice daily, frusemide, 40 mg. daily, and oxygen. The haemoglobin level rose to 9·9 g./100 ml. (68%), but the symptoms and signs persisted. Repeated blood, sputum, and urine cultures were negative and fungi were not isolated from the sputum. There was further deterioration of the appearance of the chest radiographs with the development of diffuse mottling throughout both lungs (Fig. 3).

On 27 November prednisolone was increased to 60 mg. daily, reducing over four days to 30 mg. daily, and oxygen, 6 to 8 l./min., was given continuously

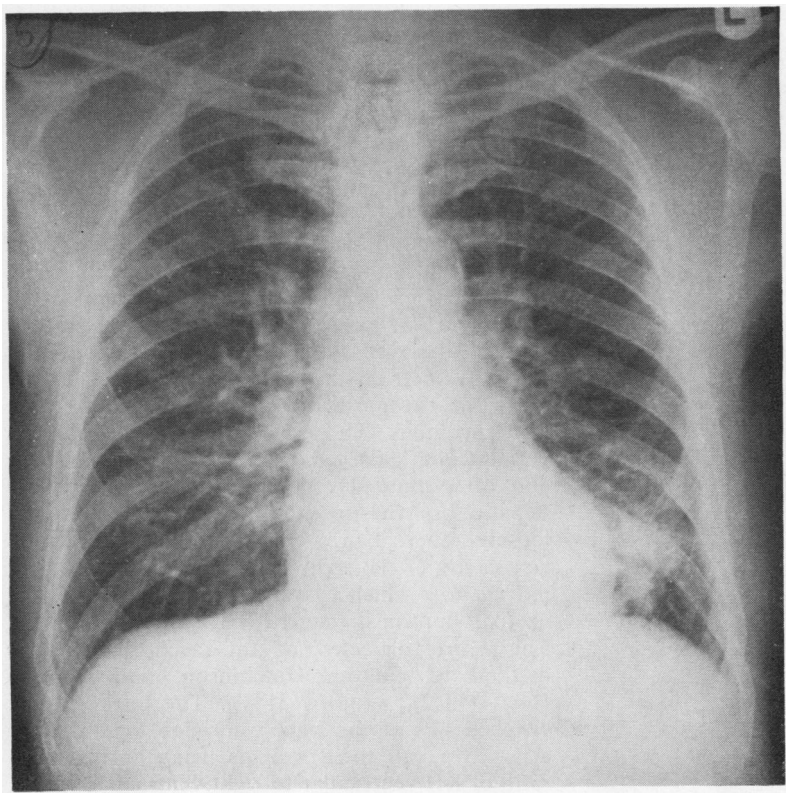

FIG. 2. *Chest radiograph taken on 6 November 1968 when the patient had had chest symptoms for two months. The radiograph shows peri-hilar infiltrates, particularly in the right sub-apical region, with an area of consolidation in the posterior segment of the left lower lobe.*

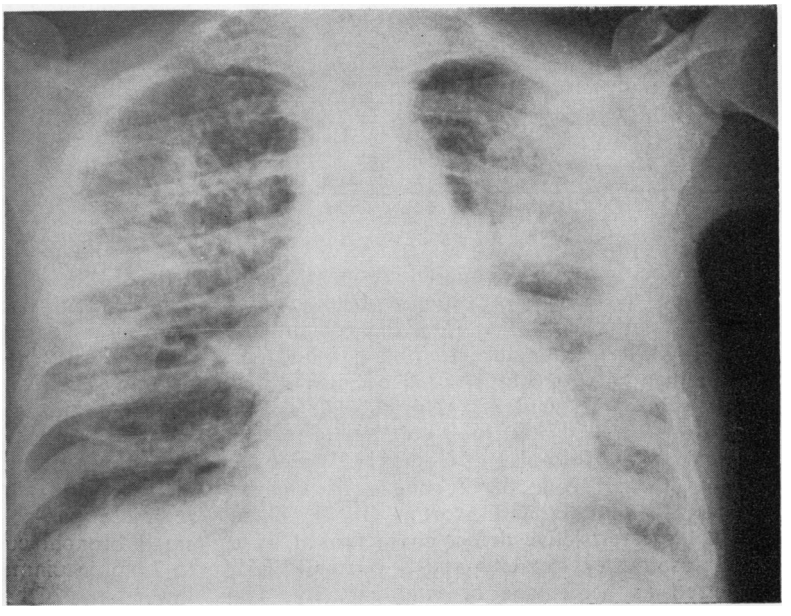

FIG. 3. *Chest radiograph taken on 6 December 1968, one week before the patient's death. There is diffuse mottling throughout both lung fields.*

by nasal catheter. Despite this treatment there was no sign of clinical or radiological improvement. Myeloblastic activity continued in the blood (on 5 December, out of a total white cell count of 16,700/cu. mm., 10% were myeloblasts, 6% myelocytes, and 8% were metamyelocytes), although the haemoglobin level remained above 70% during the last three weeks of his life. The patient died on 12 December 1968 from respiratory failure and pulmonary infection.

LUNG FUNCTION TESTS

Lung function tests were carried out on 15 November 1968 (Table). These included measurement of the ventilatory capacity, the subdivisions of lung volume, alveolar ventilation, transfer factor (by the single breath carbon monoxide method), and arterial P_{CO_2} (by the rebreathing method). Arterial puncture to obtain blood for measurement of P_{O_2} was omitted because of the patient's bleeding tendency. Normal values for ventilatory capacity and lung volumes were predicted from the formulae of Cotes (1965) and the transfer factor from the formulae of Ogilvie, Forster, Blakemore, and Morton (1957). There was a large restrictive defect characterized by a small inspiratory capacity and a normal FEV_1 expressed as percentage of vital capacity. The P_{CO_2} was reduced due to alveolar hyperventilation, and the transfer factor was grossly impaired at one quarter of the predicted normal.

TABLE

RESULTS OF LUNG FUNCTION TESTS

	Patient	Predicted Normal	% of Predicted Normal
Ventilatory capacity:			
Forced vital capacity (l.) (FVC)	1·8	3·85	47
Forced exp. volume (l. sec.) (FEV_1)	1·2	2·9	41
FEV_1/FVC (%)	67	69	97
Max. vol. ventilation: direct (l./min.)	42	126	33
Max. vol. ventilation: indirect (l./min.)	42	126	—
Lung volume (l.):			
Vital capacity	1·75	3·85	45
Inspiratory capacity	1·10	3·3	33
Exp. reserve volume	0·65	0·5	130
Functional residual capacity	2·75	2·6	106
Residual volume (RV)	2·10	2·1	100
Total lung capacity (TLC)	3·85	6·0	64
RV/TLC (%)	55	37	149
Resting ventilation:			
Tidal volume (l.)	0·9	0·4	—
Respiratory rate (per min.)	18	12	—
Minute ventilation (l./min.)	16·2	6·0–10·0	—
Alveolar ventilation (l./min.)	13·5	4·0–7·5	—
Arterial P_{CO_2} (re-breath) (mm. Hg)	32	40	—
Transfer factor (ml./mm. Hg/min.)	6·4	25·5	25

Age 61 yrs. Height 168 cm. Weight 60·3 kg.

NECROPSY FINDINGS

Necropsy was carried out three hours after death. Several of the findings were characteristic of chronic myeloid leukaemia. These included pallor and petechial haemorrhages in the skin. The *spleen* (730 g.) was enlarged and adherent to the diaphragm and greater omentum. Its cut surface showed areas of old and recent infarction. The *splenic vein* from the hilum of the spleen to the origin of the portal vein was occluded by pale firm thrombus. The *lymph nodes* were not enlarged. The enlarged *liver* (2,000 g.) was firm and of a mottled red and white colour. Intrahepatic branches of the portal vein contained loosely adherent firm grey thrombus. The medullary cavity of the right femur was filled with firm red *marrow* which contained yellowish-white foci up to 8 mm. in diameter. The *kidneys* were pale. The *heart*, dissected of fat, according to the method of Fulton, Hutchinson, and Morgan Jones (1952), weighed 315 g. The left ventricle weighed 179 g., the right ventricle 61 g., and the atria 75 g., all these weights being normal. The ratio of left ventricular to right ventricular weight was 2·9, which is normal. All the cardiac valves were normal in structure. Their circumferences were as follows: tricuspid, 11 cm.; pulmonary, 7 cm.; mitral, 9·5 cm.; and aortic, 7 cm.

Small blocks of tissue, 1 $mm.^3$, were cut from both lungs and fixed in ice-cold glutaraldehyde for electron microscopy. The lungs were then distended with 10% formalin until the pleural surfaces were smooth. After the lungs were fixed sagittal slices, 1 cm. in thickness, were cut and examined. Many areas of the lungs were pale and firm due to fibrosis which appeared to predominate in the peripheral areas of secondary lung lobules (Fig. 4). Such fibrotic change was seen predominantly in the left lung, where it formed a subpleural band of induration in the upper lobe up to 6 cm. in thickness. It proved impossible to point-count the lung, as many areas which appeared merely congested to the eye were obviously fibrotic on palpation. There was widespread centrilobular emphysema which affected all lobes and which was so severe focally as to resemble pan-acinar emphysema (Fig. 5). The small bronchi contained mucopus. Abscesses up to 2 cm. in diameter were found in the upper and lower lobes of both lungs (Fig. 6).

FIG. 4. *Cut surface of lung showing pale areas of fibrosis which are situated mainly in the peripheral areas of secondary lung lobules* (× 2).

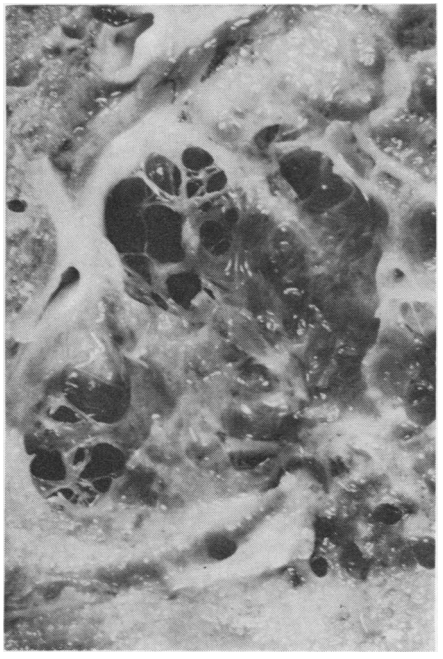

FIG. 5. *Cut surface of lung showing confluent bronchiolar emphysema of such severity as to mimic pan-acinar emphysema. There is surrounding fibrosis* (× 2).

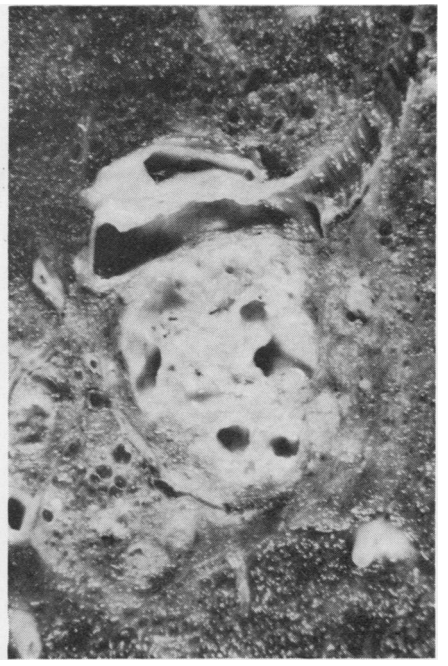

FIG. 6. *Cut surface of lung showing one of the abscesses* (× 2).

HISTOPATHOLOGY

Characteristic cells of chronic myeloid leukaemia infiltrated the spleen, the perivenous fat of the thrombosed splenic vein, the interstitial tissues of the pancreas, and the bone marrow.

LUNGS Sections of blocks of tissue taken from areas of lung not obviously fibrotic showed pronounced dilatation of alveolar capillary blood vessels with some diapedesis of red cells into alveolar spaces. In addition many alveoli contained solid masses of amorphous granular eosinophilic material (Fig. 7). While some of this may have been intra-alveolar exudate associated with the dilatation of the alveolar capillaries, much of the eosinophilic debris appeared to have arisen from the break-up of the desquamated alveolar lining cells described below (Fig. 8). In some areas of the debris there were ghost outlines of large intra-alveolar cells ; in places some of these cells were preserved relatively intact (Fig. 8). Even in the areas of lung which did not seem fibrosed to the naked eye there was early organization of this intra-alveolar debris. In the obviously fibrosed parts of the lung the organization of debris had proceeded further, with the laying down of mature fibrous tissue (Fig. 9). In many places these intra-

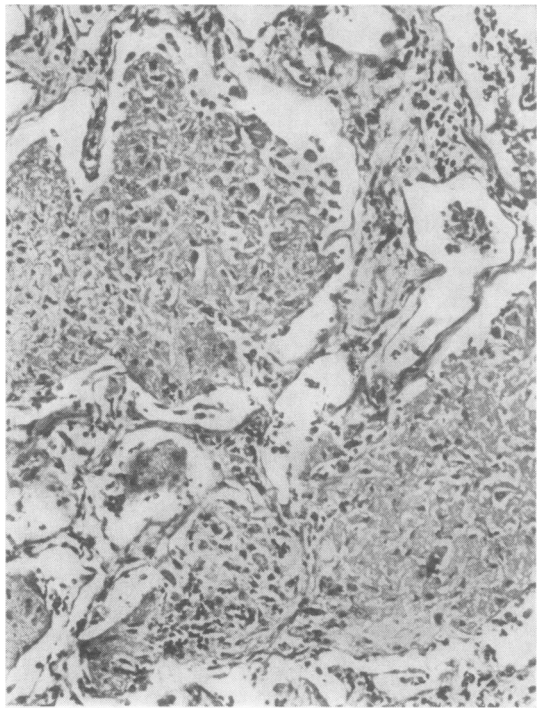

FIG. 7

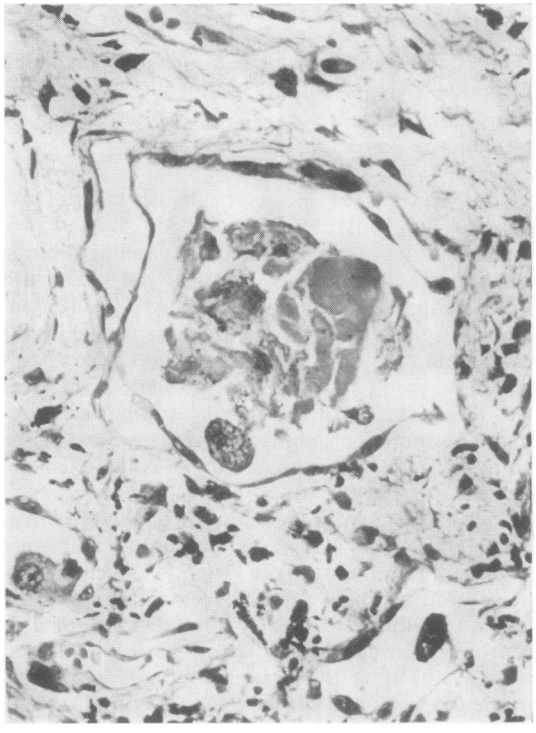

FIG. 8

alveolar masses had become adherent to and incorporated into the alveolar walls, forming fibrous nodules, some of which had coalesced to produce extensive areas of fibrosis. Many intra-alveolar masses of collagen had become covered by an extension of membranous pneumocytes from the alveolar walls. In addition to incorporation of fibrous masses on to the alveolar walls, the walls themselves showed considerable thickening due to oedema and fibrosis (Fig. 10). Thus the large fibrous areas in the lung appear to have arisen from both true fibrous thickening of alveolar walls and incorporation into them of organized masses of intra-alveolar fibrinous exudate.

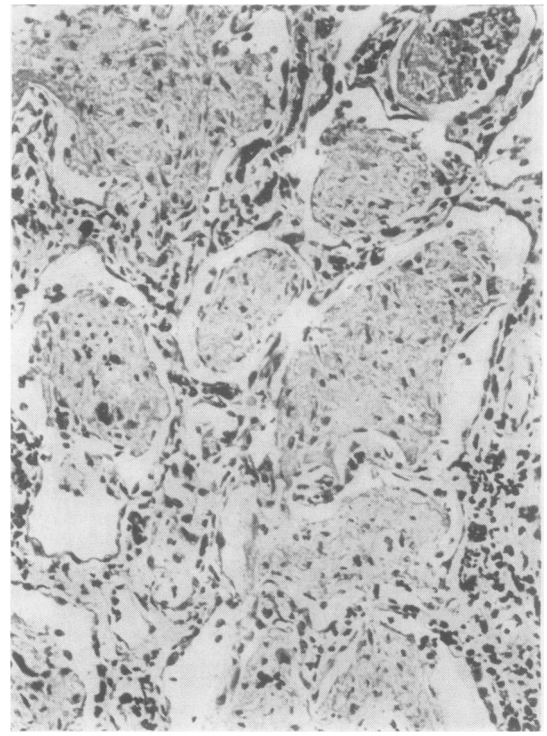

FIG. 9

FIG. 7. *Photomicrograph showing granular eosinophilic material within the alveolar spaces* (*H. and E.* × 125).

FIG. 8. *Photomicrograph showing an alveolar space which contains granular pneumocytes in various states of disintegration to form the granular material illustrated in Fig.* 7. *The surrounding lung tissue is fibrosed* (*H. and E.* × 280).

FIG. 9. *Photomicrograph showing early organization of the intra-alveolar granular material. There is associated early interstitial pulmonary fibrosis* (*H. and E.* × 145).

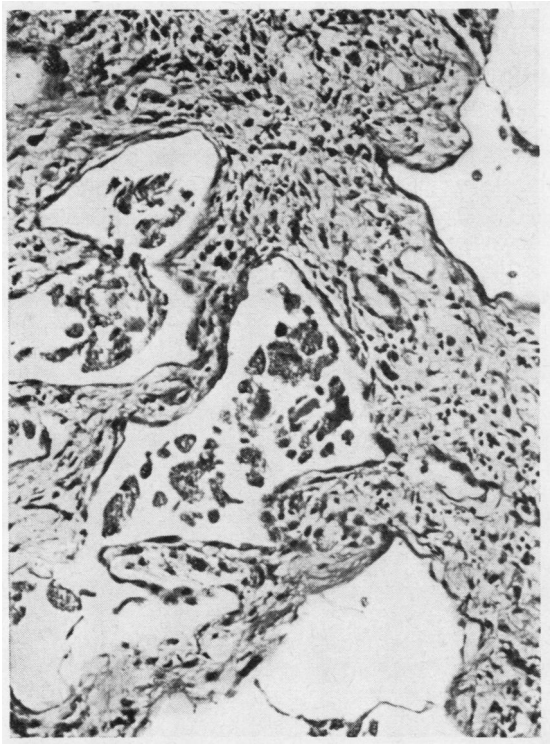

FIG. 10. *Interstitial pulmonary fibrosis with intra-alveolar collections of granular pneumocytes* (*H. and E.* × 135).

Large cells with large vesicular nuclei containing one or two prominent nucleoli were seen lining the alveolar walls (Fig. 11) or lying free within the alveolar spaces. Their cytoplasm was eosinophilic and contained numerous small clear vacuoles which gave a positive periodic acid-Schiff staining reaction. In some instances obvious microvilli were seen extending from their luminal borders. These cells, having the cytological characteristics of granular pneumocytes, projected from the alveolar walls, forming bulbous prominences and demilunes. In the alveolar spaces they either remained solitary or formed small syncytium-like masses.

The thickened alveolar walls were infiltrated by lymphocytes, eosinophils, and plasma cells. Only scanty mast cells were found in this situation, but this is not surprising since the lung had been fixed in aqueous formalin, which is known to dissolve out the metachromatic granules in human mast cells (Riley, 1959). Groups of haemosiderin-laden macrophages were present in the lung in alveolar spaces and within pleural lymphatics. These were found especially in fibrotic areas, and their presence is clearly related to the intra-alveolar haemorrhages described above.

The pulmonary vasculature showed no evidence of hypertensive pulmonary vascular disease. The muscular pulmonary arteries (100 to 1,000 μ in diameter) showed some intimal elastosis and fibrosis and there was atrophy of the underlying media related to this. The pulmonary arterioles (less than 100 μ in external diameter) had thin walls composed of a single elastic lamina and showed intimal fibrosis. A similar intimal change was seen in the small pulmonary veins and venules. Such fibrous changes in the small pulmonary blood vessels were most pronounced in and about the fibrous areas of lung. They appear to represent a combination of normal age change (Brenner, 1935) and reaction to surrounding fibrosis.

Histological examination confirmed the presence of centrilobular emphysema with dilatation of respiratory bronchioles and accumulation of carbon pigment in the surrounding lung substance. The acute abscesses noted in the lung were packed with neutrophil polymorphs and

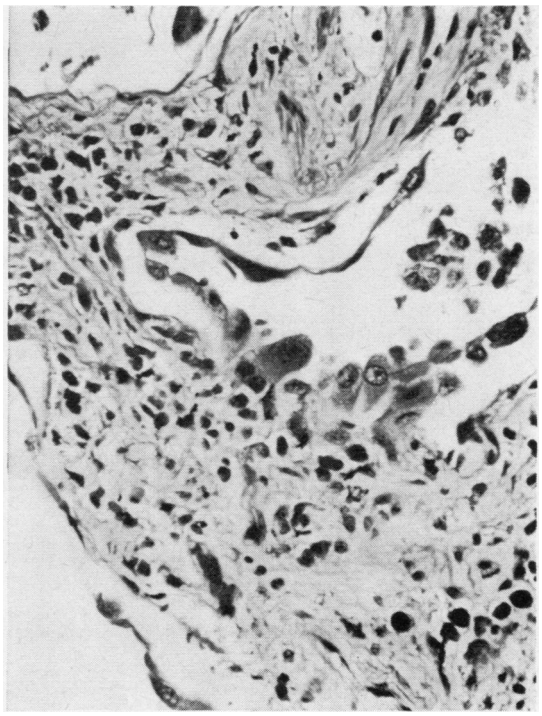

FIG. 11. *Granular pneumocytes lining an alveolar wall thickened by fibrous tissue and showing a chronic inflammatory cellular exudate* (*H. and E.* × 255).

contained fungal hyphae which stained by the periodic acid-Schiff reaction.

The pleura showed fibrous thickening and contained collateral vessels with fasciculi of longitudinal muscle in their walls.

Cholesterol granulomas were also seen. These consisted of a fibrous reaction around acicular clefts.

ELECTRON MICROSCOPY

After primary fixation with glutaraldehyde, the blocks of lung were post-fixed in osmium tetroxide, stained with uranyl acetate, and embedded in araldite. Thin sections (1 μ) were cut with an LKB Ultratome III ultramicrotome, mounted on glass slides, and stained with toluidine blue for light microscopy and the selection of suitable areas for electron microscopy. The blocks were then trimmed and ultra-thin sections (900 Å) were cut, mounted on copper grids, stained with lead citrate, and examined in an AEI EM6B electron microscope.

The enlarged alveolar epithelial cells which lined the alveolar walls (Fig. 12) and occupied the

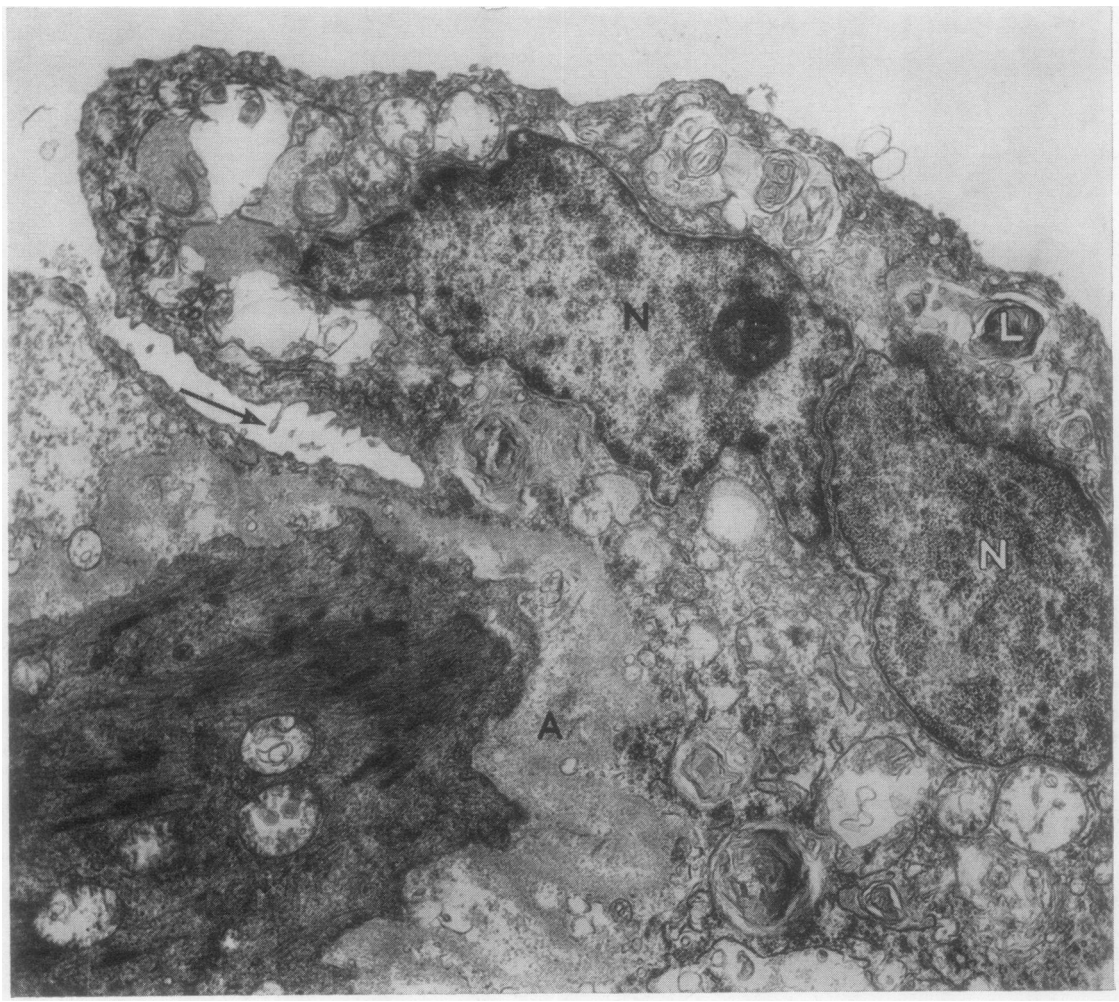

FIG. 12. *Electron micrograph showing a granular pneumocyte attached to an alveolar wall (A). The cell contains a nucleus (N) and numerous intracytoplasmic lamellar bodies (L.). Note the surface microvilli (arrow)* (× 10,270).

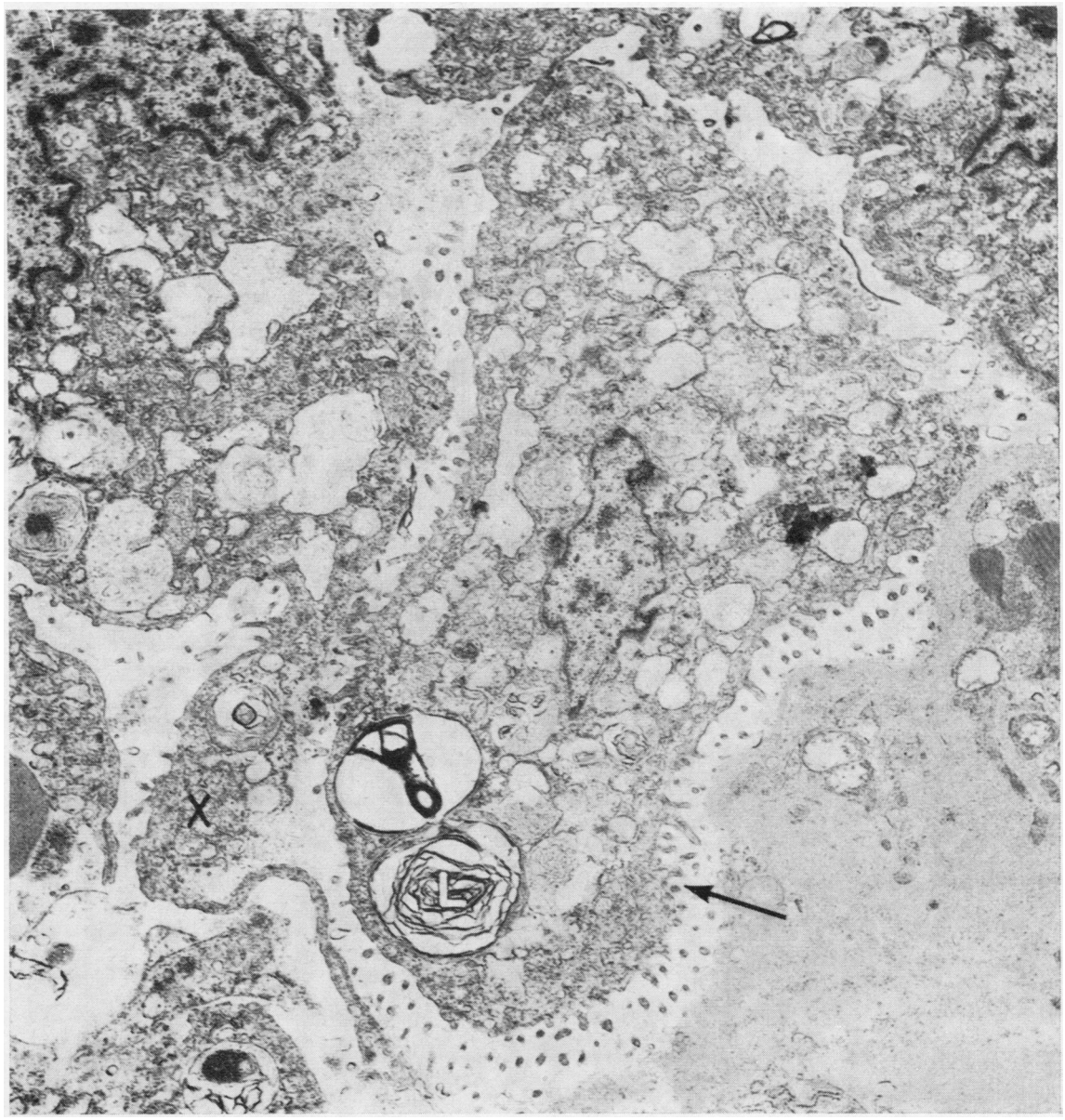

FIG. 13. *Electron micrograph showing granular pneumocytes within an alveolar space. These cells contain characteristic osmiophilic lamellar bodies (L) and have numerous short microvilli on the cell surface (arrow). Note the cytoplasmic streamer (X) of the granular pneumocyte to the left* (× 7,500).

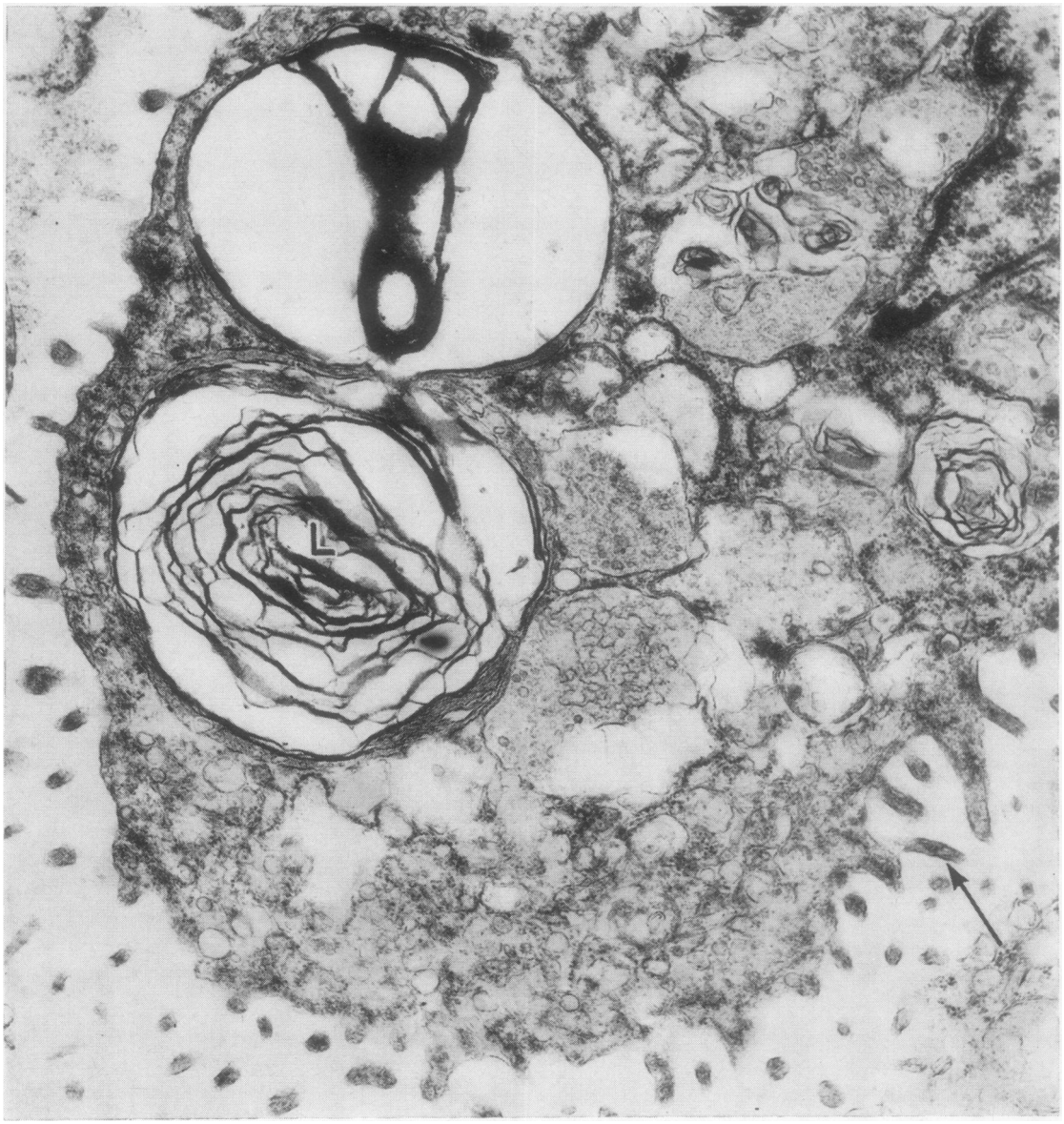

FIG. 14. *Detail of Fig.* 13 *to show the lamellar bodies* (*L*) *and microvilli* (*arrow*) (× 23,000).

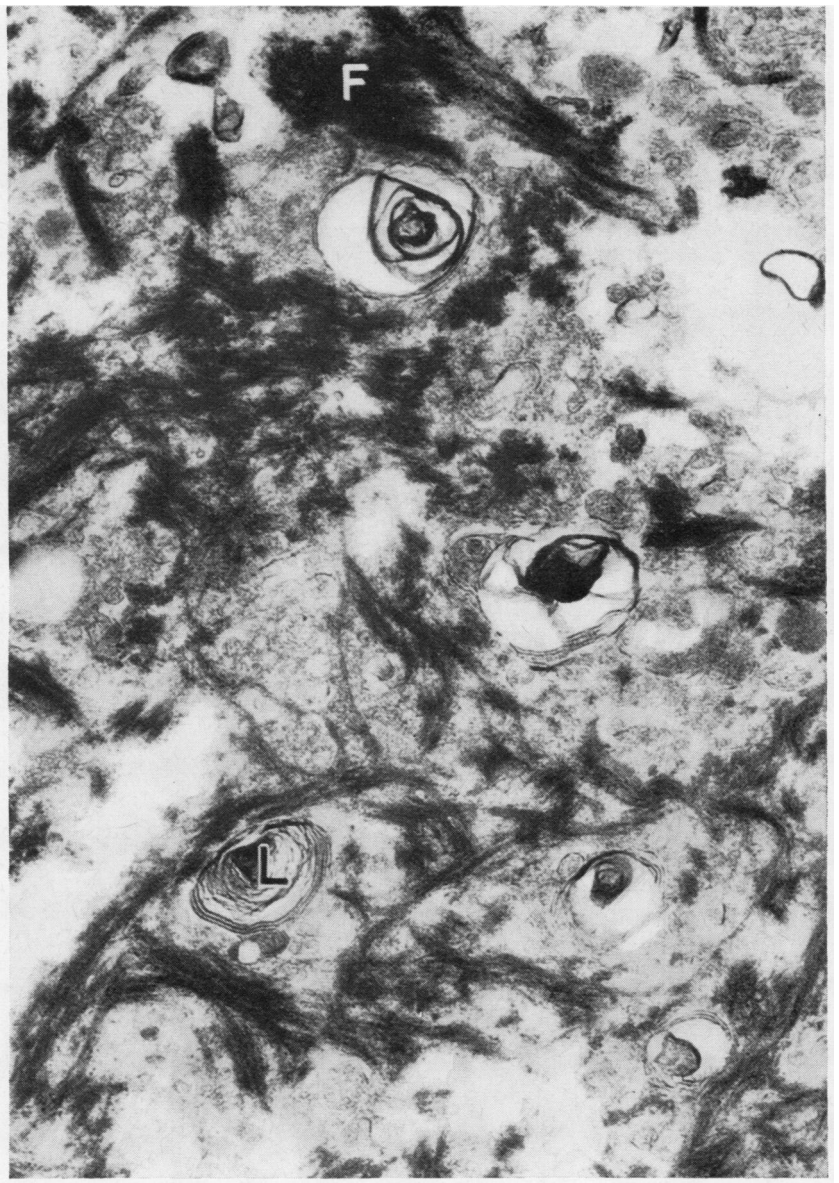

FIG. 15. *Electron micrograph showing the nature of the alveolar contents in busulphan lung. The dark strands are fibrin (F). The rounded structures, consisting of concentric osmiophilic rings, are discharged lamellar bodies (L)* (× 18,750).

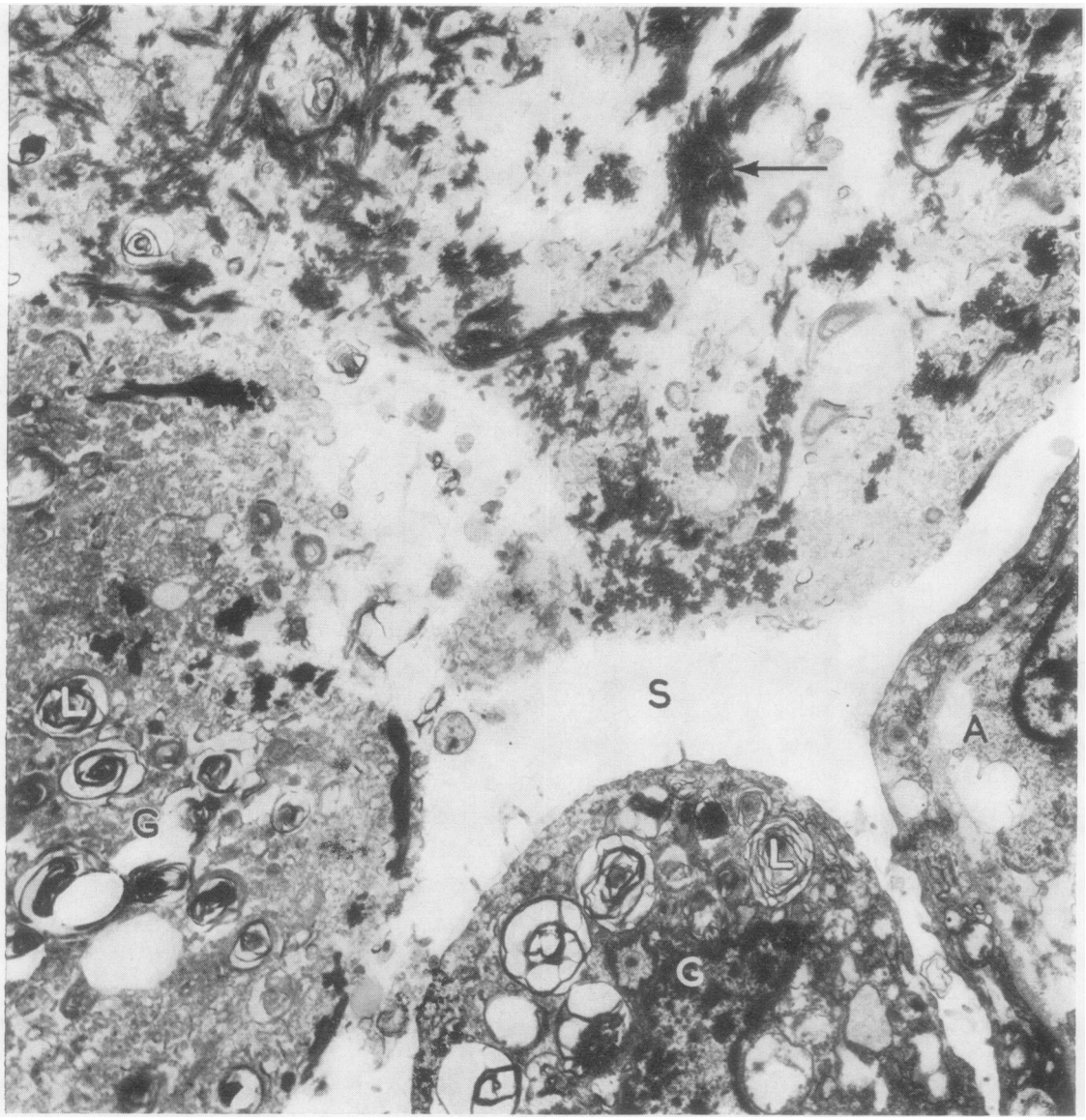

FIG. 16. *Electron micrograph showing two granular pneumocytes (G) in an alveolar space (S). Both cells contain characteristic osmiophilic lamellar bodies (L). The granular pneumocyte seen to the left has ruptured, liberating its cell contents, including lamellar bodies, into the alveolus. The dark fibrillar material (arrow) is composed of strands of fibrin. Alveolar wall (A) on right* (× 6,390).

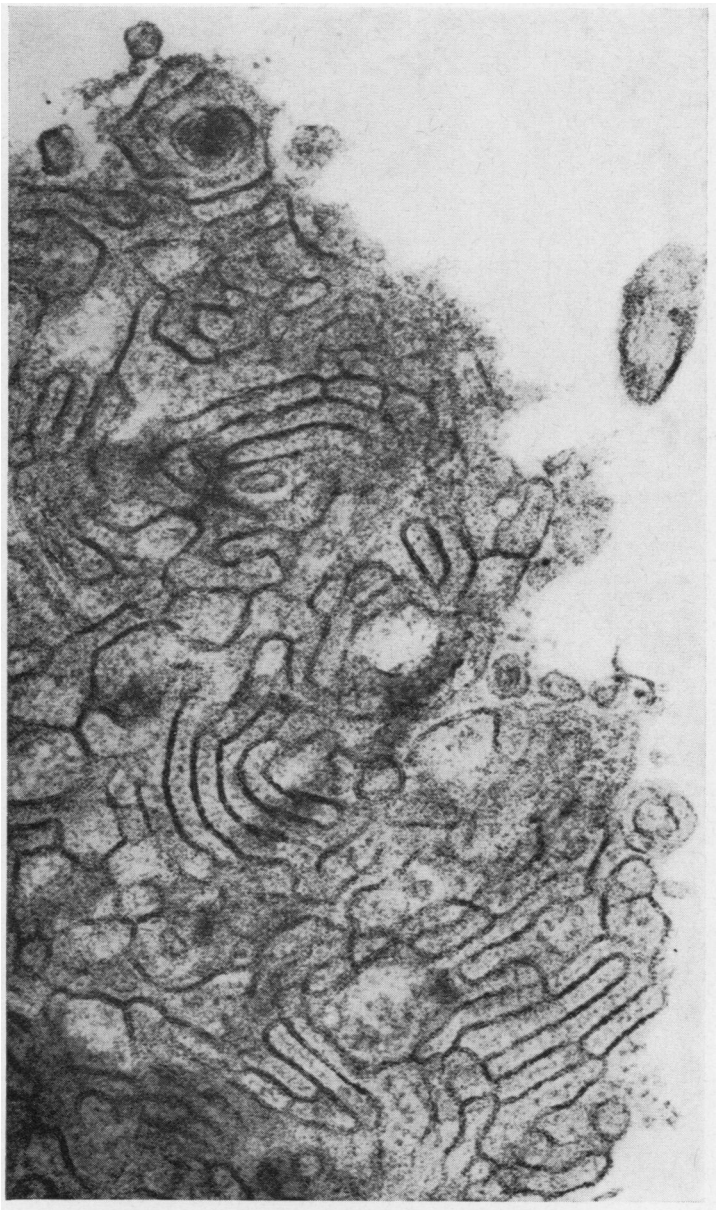

FIG. 17. *Detail of phospholipid lattice seen within an alveolar space* (× 100,000).

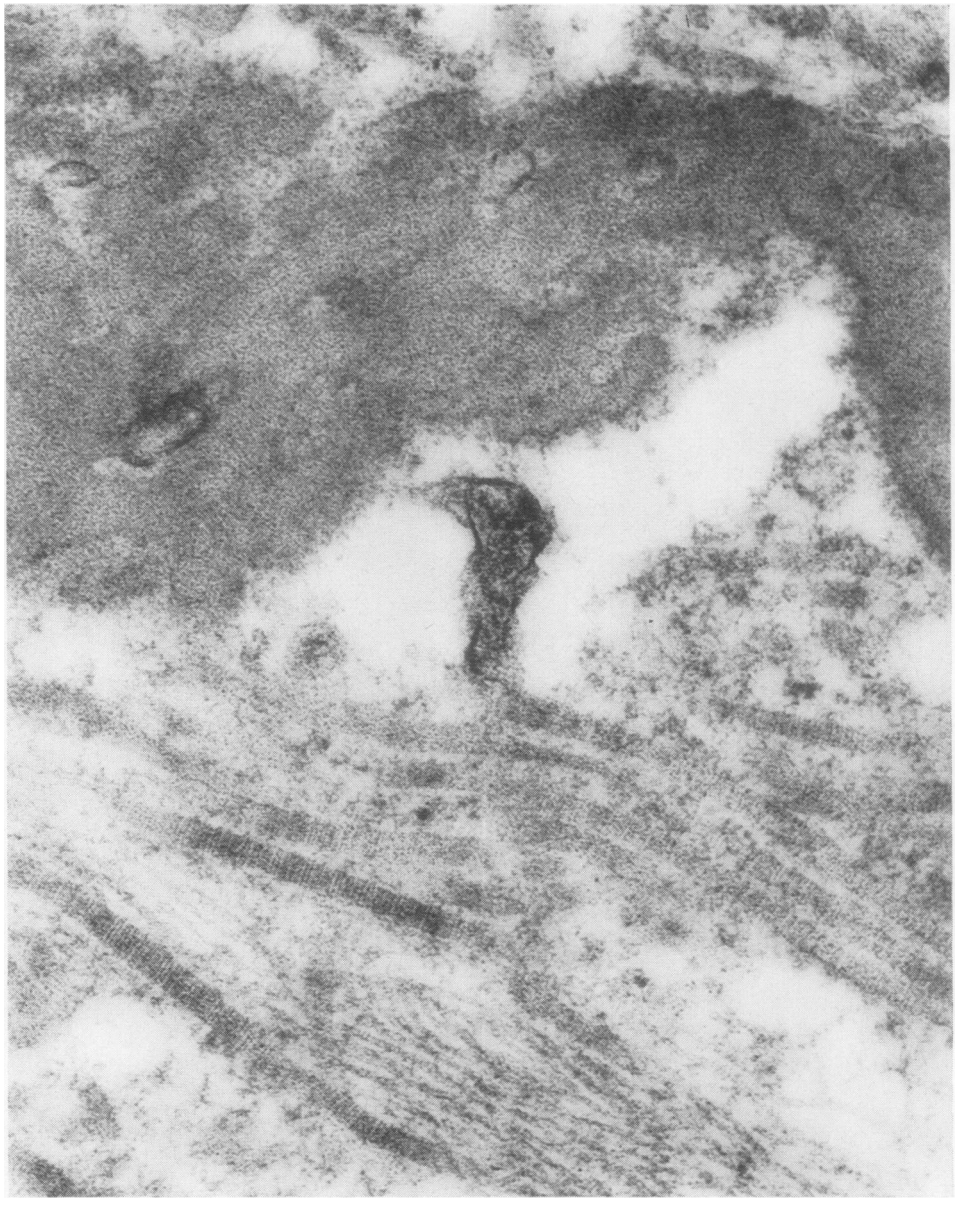

FIG. 18. *Electron micrograph to show the nature of a non-cellular component of the intra-alveolar material. The dark granular material above is probably fibrin. Below are collagen fibres showing characteristic periodicity* (× 75,000).

alveolar spaces (Fig. 13) were identified as granular pneumocytes. They possessed prominent intracytoplasmic lamellar or spiral osmiophilic secretory inclusions which are usually called 'lamellar bodies' (Fig. 14). The cell surface was thrown up into characteristic irregular, short microvilli.

The intra-alveolar debris consisted of strands of amorphous osmiophilic fibrillary material interspersed with roughly circular bodies composed of concentric osmiophilic membranes (Fig. 15). The fibrillary material resembled fibrin and the circular bodies appeared identical to the lamellar secretory inclusion bodies of granular pneumocytes. In some instances it appeared that granular pneumocytes had disintegrated (Fig. 16) and liberated their lamellar bodies, so giving rise to the intra-alveolar debris which characterized the histological picture of this condition. Also scattered within the alveolar spaces were complex lattice structures (Fig. 17) composed of osmiophilic membranes which resembled the artificial phospholipid membranes prepared by Lucy and Glauert (1964). Evidence of organization of the intra-alveolar debris was seen in the form of groups of fibrils showing the periodic transverse striations characteristic of collagen (Fig. 18).

DISCUSSION

The development of pulmonary complications in some patients on busulphan therapy has been recognized since the original report of Oliner, Schwartz, Rubio, and Dameshek in 1961. They described two patients with chronic myeloid leukaemia receiving the drug who developed dyspnoea, fever, weakness, and weight loss; one of these patients had a dry cough. The main physical findings noted, as in the present case, were bilateral basal crepitations.

Radiographs of the chest in their cases showed diffuse, bilateral infiltrates which had the appearance of an inflammatory process. From our pathological findings, and by analogy with the radiographic features of desquamative interstitial pneumonia (Liebow, Steer and Billingsley, 1965), in which there is a similar pathology of initial exudation of granular pneumocytes followed by the development of fibrosing alveolitis, we think that two radiographic pictures are to be expected in busulphan lung. In the early stage, when the alveolar spaces are filled with large numbers of granular pneumocytes, the chest radiographs should show a 'ground-glass' appearance, as in the cases of desquamative interstitial pneumonia illustrated by Liebow *et al.* (1965). In the later stage of fibrous thickening of alveolar walls and organization of intra-alveolar exudate, we would anticipate seeing the well-known radiological appearances of interstitial pulmonary fibrosis.

The radiographic abnormalities of busulphan lung were initially misdiagnosed as miliary tuberculosis by Leake, Smith, and Woodliff (1963) in one of their two cases, and this patient was given anti-tuberculosis therapy. The chest radiographs were said to be consistent with interstitial pulmonary fibrosis in the third case of Kyle, Schwartz, Oliner, and Dameshek (1961) and in the case of Bates and Christie (1964). Our case is atypical in that, while in most previously reported examples of this disease chest radiographs have usually shown changes from the onset of symptoms, in our patient they showed no abnormality in the early stages of the illness. When changes did appear eventually they were not at first characteristic, since they showed consolidation of the left lower lobe. This change appears to be related to the abscess formation with surrounding inflammatory change in this area that we noted at necropsy.

Pulmonary function tests were performed on one of the patients of Oliner *et al.* (1961) and were reported as being 'compatible with the syndrome of alveolar-capillary block', although no actual figure for the transfer factor was given. Our patient showed evidence of 'restrictive lung disease' from pulmonary function studies, with a severe reduction in transfer factor, so that it was only 25% of the normal (Table). We believe that the transfer factor may prove to be a useful indicator of the onset and severity of pulmonary complications of busulphan therapy.

Pulmonary function tests were also said to indicate 'alveolar capillary block' in one of four cases (case 3) of an Addisonian-like syndrome following busulphan therapy reported by Kyle *et al.* (1961); once again no precise data from these tests were given by this author. Similar tests carried out on one of the cases of Leake *et al.* (1963) showed a slight defect in diffusion', but no figures were given. Bates and Christie (1964) demonstrated a low transfer factor associated with a low haemoglobin during the course of illness in a patient with busulphan lung. Correction of the patient's anaemia restored the transfer factor to the normal range initially, but in the terminal stage of the illness the transfer factor was low despite an adequate level of haemoglobin. The authors drew attention to the difficulties of interpreting changes in the value of the transfer factor in the presence of anaemia. In

our patient the haemoglobin at the time that pulmonary function tests were carried out was 11·2 g./100 ml. (77%). Anaemia of this degree would not contribute significantly to the profound reduction in transfer factor which we found.

When pulmonary complications developed in their patients, Oliner *et al.* (1961) discontinued the busulphan therapy and gave large doses of prednisolone. Both patients responded to this treatment with disappearance of their pulmonary signs and symptoms; one of the patients also showed radiological improvement. It seems likely to us that improvement following the use of steroids will only occur when the disease is in its cellular phase with the exudation of granular pneumocytes. There seems little likelihood of clinical or radiological improvement with such treatment when extensive pulmonary fibrosis has occurred. The patient designated case 3 by Kyle *et al.* (1961) responded to the stopping of busulphan and the administration of prednisolone. Under this regime the pulmonary symptoms disappeared, although the patient subsequently died from pulmonary oedema. One of the patients of Leake *et al.* (1963) improved with cessation of treatment by busulphan alone; in this instance no steroids were given. Our patient was not given prednisolone initially, and when it was started it was given in relatively small doses because of haemolysis. When eventually bigger doses were employed the pulmonary syndrome was in an advanced state. Where patients have responded to prednisolone, big doses have been used (Oliner *et al.*, 1961; Kyle *et al.*, 1961) whilst in those cases where smaller doses were used (Smalley and Wall, 1966) the patients have died with very little response.

Death from pulmonary oedema has been reported in patients with busulphan lung by Oliner *et al.* (1961) and Kyle *et al.* (1961).

Our patient shows a notable difference from those reported previously in that busulphan had been stopped for one month before the onset of symptoms. In the previous cases reported the patients were taking the drug at the time when symptoms appeared. Central cyanosis was prominent in our case, but it is mentioned as being present in only one of the cases reported before (Smalley and Wall, 1966).

In the majority of cases of busulphan lung previously reported the biopsy specimens of lung have been described as showing interstitial pulmonary fibrosis (as in the cases of Oliner *et al.* (1961), the third case of Kyle *et al.* (1961), and the second case of Leake *et al.* (1963)). The concept of the pathology of this disease was broadened by the recognition of atypical intra-alveolar cells by Heard and Cooke (1968). We have been able to confirm by means of electron microscopy their suggestion made on the basis of light microscopy that these cells are the large alveolar (type II) cells. The ultrastructure of these cells in the present case is characteristic of granular pneumocytes in possessing highly characteristic lamellar bodies (Figs 13 and 15) and in showing short microvilli (Fig. 13).

We have noted the same cells in conditions characterized by chemical and physical irritation of the alveolar walls, with the production of first the cellular and later the fibrosing stages of alveolitis. Thus we have seen this proliferation of granular pneumocytes in rats poisoned by *Crotalaria spectabilis* seeds to produce pulmonary hypertension (Kay, Smith, and Heath, 1969). We have seen the same phenomenon in the lungs of dogs subjected to irradiation, sent to us for examination by Dr. H. W. C. Ward, of the Queen Elizabeth Hospital, Birmingham. Finally, we have demonstrated by electron microscopy (Brewer, Heath, and Asquith, 1969) the same outpouring of granular pneumocytes into the alveoli in a case of the non-specific cellular alveolitis which has been termed desquamative interstitial pneumonia (Liebow *et al.*, 1965). It is apparent that the granular pneumocyte is a common reactive cell in the alveolar wall in man, responding to all manner of noxious stimuli. The presence of such cells is in no way specific for busulphan lung.

We suggest that the other features of the pathology of busulphan lung follow this cellular alveolitis and we differ from Heard and Cooke (1968) in their interpretation of the histopathology. They are of the opinion that the intra-alveolar eosinophilic material is to be explained on the basis of a persistent fibrinous oedema. We think it likely that much of this intra-alveolar substance represents the breakdown of previously desquamated granular pneumocytes. We have arrived at this conclusion by noting the intimate admixture of intact or ghost outlines of pneumocytes with the intra-alveolar debris (Figs 8 and 16). Fibrinous oedema is associated in the production of this alveolar material, but the disintegration of granular pneumocytes also plays an important role.

We agree with Heard and Cooke (1968) that much of this material undergoes subsequent organization and incorporation into alveolar

walls, but we believe from our study of this case that true innate fibrous thickening of alveolar walls occurs. This appears to follow the natural history of alveolitis progressing from the cellular to the fibrous stage, without in all cases an associated thickening of alveolar walls brought about by organization and incorporation of intra-alveolar material.

It seems likely, too, that the disintegration of the granular pneumocytes (Fig. 16), with liberation of the phospholipid contained in their lamellar bodies (Figs 15 and 16), may be responsible for the lipid granulomas noted in this case. Glancy, Frazier, and Roberts (1968) describe cholesterol granulomas of identical histological structure which they were able to show by *x*-ray diffraction studies contained cholesterol palmitate and stearate. Although these authors associated such granulomas with pulmonary hypertension, it should be noted that identical granulomas in this case occurred without any morbid anatomical evidence of a raised pulmonary arterial pressure. It seems to us more likely that granulomas of this type arise as a reaction to phospholipid liberation by disintegrating granular pneumocytes.

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