Microanalyses of lesions and lymph nodes from coalminers’ lungs

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ABSTRACT  The dust content and composition of lesions and hilar lymph nodes from the lungs of British coalworkers have been examined. Samples of macules, fibrotic nodules, and massive fibrosis (both peripheral and central sites) were dissected from 49 lungs. The highest mean dust concentrations (about 20%) were found in nodules and massive fibrosis. Overall there were no significant differences between the selected lesion types and their respective whole lung dust composition, although the central sites of massive fibrosis were found to contain on average a higher proportion of coal and a lower proportion of ash and its measured constituents, quartz and kaolin plus mica, than the edge of the lesion (p < 0.001 for each component). There were striking differences between recovered lung and lymph node dusts. An examination of 180 specimens showed a mean quartz in lymph node dust of 20.3% compared with 6.1% in lung dust. As expected the proportion of quartz was greater in lymph nodes and lungs from men who had worked “low” rank (high ash) coal. By contrast with the corresponding figures for lung dusts, however, the mean proportion of quartz in nodes did not increase over the pathological range of pneumoconiotic lung disease. On average the proportions of kaolin and mica in lymph nodes reflect those found in lungs. The lymphotrophic nature of quartz was clearly shown although it was not possible to show an association between this clearance pathway and any particular type of lesion.

Pathological and mineralogical studies of lungs from British coalminers employed at collieries included in the National Coal Board’s Pneumoconiosis Field Research have been reported by Davis et al1 and Ruckley et al.2 They showed that, on average, the mass and composition of dust retained in lungs varied according to both the rank of coal mined and the extent of pathological response to that dust. In general, subjects showing progressive massive fibrosis (PMF) retained more dust per unit increase in dust exposure than those showing less severe disease. For some, particularly those with pneumoconiosis who had worked low rank coal, the proportion of ash components in lung dust was greater than would be expected from the composition of respirable dust. Wagner et al found that, within a lung, coal content of tissue was greatest in the centre of an area of massive fibrosis.3 In addition there is experimental evidence that quartz is preferentially transferred to lymph nodes.4-6

We therefore considered it desirable to examine the dust content and composition of lymph nodes and of the various lesions found within an individual lung since such analyses may indicate how the disease developed and suggest clearance mechanisms.

Materials and methods

SELECTION OF LUNGS
The original necropsy study included 500 sets of lungs with a measure of lung dust content available for 490 of these specimens. In the course of the study one lung from each pair was retained in store, the other having been used in determining the overall dust burden.

Forty nine of the retained lungs were chosen for the present work because they contained a sufficient number and variety of lesions for examination. Care was taken to include examples of exposure to the various ranks of coal mined.

During the latter half of the necropsy study, lymph nodes, when available, were dissected from the hilum of the lung and analysed separately. Hilar
Table 1  Recovered dust from representative whole lung samples and isolated lesions expressed as a mean percentage (with standard deviation) of the relevant dried tissue weight, together with the range of observations

<table>
<thead>
<tr>
<th>Tissue</th>
<th>No of specimens</th>
<th>Mean % dust</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Whole lung*</td>
<td>49</td>
<td>8.8 (4.2)</td>
<td>2.3-24.5</td>
</tr>
<tr>
<td>Lymph nodes</td>
<td>31</td>
<td>11.4 (4.1)</td>
<td>4.6-21.1</td>
</tr>
<tr>
<td>Macules</td>
<td>48</td>
<td>15.4 (13.1)</td>
<td>2.0-85.1†</td>
</tr>
<tr>
<td>Parenchymal fibrotic nodules</td>
<td>49</td>
<td>18.8 (9.9)</td>
<td>2.9-43.6</td>
</tr>
<tr>
<td>Isolated gross PMF</td>
<td>29</td>
<td>20.2 (12.6)</td>
<td>4.9-49.7</td>
</tr>
<tr>
<td>PMF: peripheral</td>
<td>29</td>
<td>23.7 (12.3)</td>
<td>8.6-49.2</td>
</tr>
<tr>
<td>PMF: central</td>
<td>14</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*Recovered whole lung dust from opposite side to that for dissected lesions.
†Next highest percentage for a macule was 34.4.

PMF = Any nodular lesion of 10 mm or greater in diameter.

nodes were examined from 180 specimens, of which 31 were included in the group of 49 used to study lesion types.

LESION TYPES AND SAMPLING

The types of dusted tissue examined were:

Macules—Circumscribed dusted areas offering no resistance to palpation.

Nodules—Palpable lesions of 1 mm < 10 mm in diameter. Samples were taken throughout the lung including subpleural sites when available.

PMF—Any nodular lesion of 10 mm or greater in diameter. When distinguishable, peripheral (PMF P) and central (PMF C) portions of the lesion were sampled.

Lymph nodes—Dissected from the hilum of the lung but were not from a specific anatomical site, nor did they include all nodes. The material available depended on handling before receipt at the institute.

Examples of each lesion type were dissected so that the minimum of extraneous tissue was included. The number of lungs sampled for a particular type of lesion was determined by the availability of that type in the individual lung. For example, the lung containing a unilateral PMF lesion may have been used for a whole lung dust analysis; no PMF specimen can then be obtained.

For the examination of data relating to dust in lymph nodes the lungs were divided into three pathological groups defined as follows:

M—Showing macules only,

F—Containing one or more nodules as defined above, and

PMF—Showing, in addition to macules and nodules, one or more lesions of 10 mm or greater in size.

In addition, specimens were divided into “high” and “low” rank groups depending on the rank of coal mined at the pit where each man had worked, the carbon content of the coal being above or below 88% carbon respectively.

DUST RECOVERY AND ANALYSIS

The lesions were drained and samples of the same type from a given specimen were bulked when necessary to obtain around 30 mg wet weight for analysis. After drying at 100°C for three days dust was recovered by a method in which the tissue is removed by hydrolysis with 11.3 M hydrochloric acid at 60°C. The method was adapted for collection of micro quantities of residue by filtration on to Nucleopore polycarbonate membrane filters (0.4 μm pore size). All reagents were filtered before use. The coal content of the lesions was obtained from the weight loss on ashing of the residue dusts. Quantitative analysis for quartz and kaolin plus mica in the ash was carried out by infrared spectrophotometry. Several dust samples were prepared by low temperature plasma ashing to confirm that no dehydration of the kaolins had taken place during this step. All the analyses were duplicated.

Results

DUST BURDEN

Table 1 shows the number of specimens examined for each type of lesion with the mean and range of recovered dust expressed as a percentage of dried...
Microanalyses of lesions and lymph nodes from coalminers' lungs

Table 3  Comparison of the dust composition of peripheral and central samples of PMF lesions for 14 specimens

<table>
<thead>
<tr>
<th>% Coal in recovered dust</th>
<th>% Quartz in recovered dust</th>
<th>% Kaolin plus mica in recovered dust</th>
</tr>
</thead>
<tbody>
<tr>
<td>PMF P</td>
<td>PMF C</td>
<td>PMF P</td>
</tr>
<tr>
<td>39-0</td>
<td>43-2</td>
<td>9-8</td>
</tr>
<tr>
<td>85-5</td>
<td>85-4</td>
<td>2-2</td>
</tr>
<tr>
<td>86-8</td>
<td>86-0</td>
<td>2-2</td>
</tr>
<tr>
<td>67-8</td>
<td>71-4</td>
<td>5-6</td>
</tr>
<tr>
<td>72-3</td>
<td>76-6</td>
<td>4-7</td>
</tr>
<tr>
<td>62-6</td>
<td>65-0</td>
<td>7-3</td>
</tr>
<tr>
<td>20-1</td>
<td>24-8</td>
<td>16-2</td>
</tr>
<tr>
<td>31-7</td>
<td>38-0</td>
<td>14-6</td>
</tr>
<tr>
<td>22-3</td>
<td>27-9</td>
<td>16-0</td>
</tr>
<tr>
<td>27-4</td>
<td>36-8</td>
<td>9-4</td>
</tr>
<tr>
<td>28-3</td>
<td>36-4</td>
<td>10-2</td>
</tr>
<tr>
<td>22-8</td>
<td>25-5</td>
<td>17-6</td>
</tr>
<tr>
<td>55-7</td>
<td>54-6</td>
<td>12-6</td>
</tr>
<tr>
<td>Mean</td>
<td>49-2</td>
<td>9-6</td>
</tr>
<tr>
<td>(Standard deviation)</td>
<td>(24-4)</td>
<td>(5-2)</td>
</tr>
</tbody>
</table>

PMF P = Peripheral sample.  
PMF C = Central sample.

tissue weight. The dust concentration trend evident for the mean values was followed by 30 of the 49 subjects.

Nodules and PMF showed the highest mean dust concentrations at around 20% of tissue weight and dust concentration was least in lymph nodes at 11% of tissue weight. No difference was shown between upper and lower lobe lesions examined for macules in eight subjects and for nodules in seven subjects.

Differences were observed between peripheral and central areas of PMF such that the central area contained significantly more dust per unit of dried tissue (p < 0.05, analysis of variance).

DUST COMPOSITION

On average, the coal, quartz, and kaolin plus mica present in each type of lung sample accounted for 95% of the recovered dust. There were no significant differences between the mean proportions of coal, quartz, and kaolin plus mica for any type of lesion and the whole lung dust composition of the relevant specimens (table 2). No difference was shown between upper and lower lobe lesions of the same type.

For 11 of the 14 PMF specimens, which were divided into peripheral and central areas, the central area contained a higher proportion of coal and lower proportion of ash and its measured constituents, quartz and kaolin plus mica, than the edge of the lesion (p < 0.001 for each component, analysis of variance) (table 3).

A striking difference was noted between whole lung dust and lymph node dust such that the proportion of coal was lower and that of quartz appreciably higher in lymph nodes than in the lung (table 2). These observations were explored further in the larger group of 180 specimens from which lymph nodes were dissected. The results reflect those found for 31 subjects in the lesion study. Thus the mean proportion of coal in dust from 180 lymph nodes was 48.6% whereas the corresponding lung dust showed 63.8% coal. The proportion of quartz in

Table 4  Mean percentage composition of dust recovered from whole lungs and associated lymph nodes for 180 subjects, divided by pathological group and by rank of coal mined. (Standard deviation given in parentheses)

<table>
<thead>
<tr>
<th>Coal rank</th>
<th>Pathological group</th>
<th>No of subjects</th>
<th>% Coal Total lung</th>
<th>% Quartz Total lung</th>
<th>% Kaolin + mica Total lung</th>
</tr>
</thead>
<tbody>
<tr>
<td>High</td>
<td>M</td>
<td>17</td>
<td>18-2 (12-5)</td>
<td>18-6 (7-7)</td>
<td>15-2 (8-0)</td>
</tr>
<tr>
<td></td>
<td>F</td>
<td>40</td>
<td>18-4 (11-4)</td>
<td>18-6 (7-9)</td>
<td>20-1 (8-3)</td>
</tr>
<tr>
<td></td>
<td>PMF</td>
<td>44</td>
<td>18-2 (15-4)</td>
<td>16-5 (10-0)</td>
<td>21-0 (11-4)</td>
</tr>
<tr>
<td></td>
<td>Overall</td>
<td>101</td>
<td>18-2 (13-2)</td>
<td>17-7 (8-8)</td>
<td>19-2 (9-8)</td>
</tr>
<tr>
<td>Low</td>
<td>M</td>
<td>13</td>
<td>18-2 (20-6)</td>
<td>18-6 (14-8)</td>
<td>24-1 (15-6)</td>
</tr>
<tr>
<td></td>
<td>F</td>
<td>33</td>
<td>18-2 (15-5)</td>
<td>25-2 (9-3)</td>
<td>32-0 (11-1)</td>
</tr>
<tr>
<td></td>
<td>PMF</td>
<td>33</td>
<td>18-2 (16-5)</td>
<td>21-4 (8-1)</td>
<td>37-0 (12-0)</td>
</tr>
<tr>
<td></td>
<td>Overall</td>
<td>79</td>
<td>18-2 (18-2)</td>
<td>23-5 (10-0)</td>
<td>32-8 (13-0)</td>
</tr>
</tbody>
</table>

M = Showing macules only.
F = Containing one or more nodules (see text).
PMF = Any nodular lesion of 10 mm or greater in diameter.
lymph node dust was 20.3% compared with 6.1% in whole lungs; kaolin plus mica was present in similar proportions in lymph node and lung dust at around 26%. The composition of dust retained in lymph nodes showed the expected pronounced differences between the two broad “rank” groups (table 4). Nevertheless, the observed differences in lung dust composition over the pathological range for low rank subjects was not seen in the corresponding values for lymph node dust except for kaolin plus mica which, in both rank groups, closely reflects the proportions seen in whole lung dust.

The greater proportion of ash in lymph node dust compared with that in the lungs was therefore almost wholly accounted for by a much higher proportion of quartz in lymph nodes (table 4).

Comment

In the present study virtually all the men showed the full range of pathological responses to dust in their lungs. The dust concentration trend observed for the various types of lesions analysed in this study is as might be expected. The pattern of greater dust concentration in massive fibrosis and fibrotic nodules when compared with macules also reflects the often diffuse nature of the latter which on dissection would have been expected to include relatively high amounts of associated tissue. Other work, however, suggests that simple dust accumulation cannot explain the wide range of pathological abnormality which may be found for a given lung dust burden. Several factors are implicated in the pathogenesis of PMF, of which two candidates are the composition of the dust to which the men were exposed and the tissue reaction to that dust.

Compositional differences in the recovered lung dust of men from different mining areas are well documented.12910 To a large extent they reflect differences in the dust to which men have been exposed, although it has been observed that retained lung dust may be enriched in terms of ash components or depleted of coal, when compared with respirable dust.1112 Davis et al showed this as being a feature of low rank subjects with established pneumoconiosis.1 This difference in dust composition may be due to the removal of the less toxic material, coal, because it is not fixed in tissue by a strongly fibrotic reaction. Conversely, there may be preferential deposition of ash components because they elicit a more vigorous cellular response that favours their retention in the lung or removal by the lymphatic system.

The lymphotrophic nature of quartz has been extensively shown in animal work, most recently by Reisner et al5 and Robertson et al.6 The present results confirm this in a large group of coalworkers showing, for all grades of pathological lung involvement, a much higher proportion of quartz in lymph node dust than in lung dust. Although accounting for only a small part of the dust clearance in mass terms, we thought that disturbance of this pathway might make a disproportionate contribution to development of disease by encouraging quartz accumulation in the lung. This led us to consider whether a pattern of quartz clearance within the lung could be identified. In this respect the overall results were disappointing. We found no significant differences in the composition of the recovered dust for any lesion type and their respective whole lung value. Perhaps a more careful determination of the anatomical site of nodules within the lung would allow distinction of those that are a response to quartz accumulation. The suggestion that the lymphatic mechanism might be disturbed is not new13 and would be particularly applicable in men exposed to the greatest amount of ash in respirable dust. Although disputing that true obstruction might occur within the lymphatic system, Heppleston concluded that the presence of dust foci might impede entry to the lymphatics of dust subsequently deposited.14

In the present study a close positive association between lung and lymph node dust was shown for percentage coal, ash, and kaolin plus mica (r = 0.8). A similar association for percentage quartz was less striking (r = 0.4); we hope that investigation of this relationship with reference to extent of pathological change and rank of coal will help to elucidate the role of quartz in disease development.

We thank the staff at the Institute of Occupational Medicine for their skilled help and acknowledge gratefully the financial support of the National Coal Board and the Commission of the European Communities.

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