Original Article

Anthracotic Pigment in Transbronchial Lung Biopsy: An Innocent Bystander or Pathogenic Agent for Parenchymal Lung Disease

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Abstract

Background. Anthracosis has been recently identified as a cause of bronchitis and bronchial stenosis in both developing and developed countries in the world. However, its exact nature whether as an innocent bystander or pathogenic agent for parenchymal lung disease is unknown.

Methods. We retrospectively analysed 384 transbronchial lung biopsies (TBLBs) received at Department of Pathology over a seven-year period (August 2010 to August 2016). Thirteen TBLBs showed normal lung parenchyma were taken as controls; 32 (8.3%) TBLBs showed deposition of anthracotic pigment, with or without fibrosis and were further studied. Masson-Trichrome and Ziehl-Neelsen stains were used to confirm the diagnosis of fibrosis and tuberculosis, respectively.

Results. The TBLBs were histopathologically categorised into: Group 1 normal lung parenchyma, (controls, n=13, 3.4%); Group 2: pigment deposition with fibrotic parenchymal reaction (n=11, 34.4%); Group 3: pigment deposition with inflammatory parenchymal reaction (n=11, 34.4%); and Group 4: pigment deposition with granulomatous parenchymal reaction (n=10, 31.3%). In two cases of Group 2 and one case of Group 3, parenchymal deposits of silicate crystals were also identified by polarising microscopy.

Conclusions. Anthracosis does not appear to be an innocent bystander and needs to be meticulously assessed for its role as pathogenic agent for parenchymal lung disease in all cases. Our observations suggest that identifying the pigment deposited and correlation with the underlying pathology in the limited tissue sample available can help in reaching a definitive diagnosis. **[Indian J Chest Dis Allied Sci 2018;60:27-31]**

Key words: Anthracosis, Bronchial anthracofibrosis, Granulomatous inflammation, Transbronchial lung biopsy.

Introduction

Anthracosis is the deposition of coal/carbon particles and other black pigments in the lungs. It is found in heavy smokers, in city dwellers exposed to high environmental particulate matter and after occupational exposures to coal. This condition has been traditionally associated with pneumoconioses[1] and was initially reported in mummies.[2,3] Chung *et al*,[4] retrospectively analysed the Korean patients with anthracotic pigment deposition, focal mucous fibrosis and bronchial stenosis and termed this distinct clinical entity as "bronchial anthracofibrosis". A predominance of anthracofibrosis was later identified in elderly non-smoking women with a long standing history of exposure to wood smoke used for cooking.[5]

These patients usually present with chronic cough, exertional dyspnoea and other constitutional symptoms that may be similar to bronchogenic carcinoma and endobronchial tuberculosis (TB). The bronchial narrowing with distal atelectasis seen on radiology further adds to the diagnostic dilemma. Adding to the above is the recognition that anthracotic pigment can co-exist with granulomatous infections, such as TB, malignancy and biomass fuel exposure, emphasising the need for accurate tissue diagnosis.[6] Recent studies[7,8] from India have attempted to study the association of anthracosis with demographic variables, biomass fuel, and occupational exposure. These studies revealed that 60% cases of anthracosis have been diagnosed with either old or active pulmonary TB. However, these observations are in contrast to another study[8] in which no relationship was found between anthracosis and TB. Therefore, relationship between these diseases needs to be further studied.

Transbronchial lung biopsy (TBLB) is a commonly used technique for ascertaining tissue diagnosis in pulmonary diseases. It is limited by the small size of the biopsy and the centrilobular location of the tissue obtained. While interpreting the anthracotic

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pigment seen on TBLB, the identification of the pigment and its location and associated tissue reaction needs to be correlated to the rest of the biopsy pathology to ascertain its significance. A previous study[9] had evaluated and defined to a certain extent the clinical, radiological and bronchoscopic findings of patients of anthracofibrosis. However, no study has focused on the biopsy pathology of these patients and the parenchymal tissue reaction to the same. Therefore, in the present study, we evaluated the lung parenchymal tissue reaction and anthracotic pigment deposition on TBLB.

Material and Methods

We retrospectively analysed 384 TBLBs received at Department of Pathology, Vallabhbhai Patel Chest Institute, Delhi, India from August 2010 to August 2016. Demographic data, history of exposure to smoke and associated clinical features were recorded. Most of the patients presented with cough, dyspnoea and haemoptysis and underwent bronchoscopy to rule out underlying diffuse parenchymal lung diseases.

Results

The TBLBs were histopathologically studied for the presence of anthracotic pigment deposition, presence and extent of inflammatory and fibrotic reaction in lung parenchyma and evidence of granuloma formation. Thirteen of 384 (3.4%) TBLBs showed normal lung parenchyma (Figure 1) and were taken as controls. These included nine males and four females with a mean age of 56 years (15 to 85 years). Thirty-two of 384 (8.3%) TBLBs showed parenchymal deposition of anthracotic pigment. These included 21 males and 11 females with a mean age of 56 years (24 to 88 years) (Table 1). The TBLBs were histopathologically categorised into four groups on the basis of the lung parenchymal changes: Group 1 controls (n=13); Group 2: pigment deposition with fibrotic parenchymal reaction (n=11); Group 3: pigment deposition with inflammatory parenchymal reaction (n=11); and Group 4: pigment deposition with granulomatous parenchymal reaction (n=10). In two cases in Group 2 and one case in Group 3, parenchymal deposits of silicate crystals were also identified by polarising microscopy. Masson-Trichrome and Ziehl-Neelsen stains were used to confirm the diagnosis of fibrosis and TB, respectively.

Normal lung parenchymal histopathology was seen in lung biopsies in the control group (Figure 1). No parenchymal deposition of anthracotic pigment was seen in these patients despite a history of *chullah* smoke/biomass, cigarette/*bidi* smoke and occupational dust exposure in 6 out of 13 (46.2%) (Table). The duration of smoking in controls ranged from two months to four years. No evidence of silica and/or aluminum deposit was seen in the lung biopsies of the controls when examined by polarising microscopy. In Group 2, average number of years for smoking was higher (range 35-40 years), while in Group 3, this was less (range 2-20 years) which was suggestive of an association between the deposition of anthracotic pigment and the duration of exposure (Table). In Group 2, anthracotic pigment deposition was surrounded by fibrotic parenchymal reaction that resulted in nodule formation.

Table. Demographic variables

Variable	Patients (n=32)	Controls (n=13)
Age (mean) (years)	56	56
Male	21/32 (65.6%)	9/13 (69.2%)
Female	11/32 (34.4%)	4/13 (30.8%)
History of cigarette/*bidi* smoking	10/32 (31.3%)	4/13 (30.8%)
History of *chullah* smoke/ biomass exposure	5/32 (15.6%)	1/13 (8.0%)
History of anti-tubercular therapy	5/32 (15.6%)	2/13 (15.4%)
History of occupational dust exposure	4/32 (12.5%)	1/13 (8.0%)

The collagen fiber deposition was confirmed using Masson-Trichome stain (Figure 2). In Group 3, interstitial deposits of anthracotic pigment were macular in nature and associated with inflammatory cell infiltrates leading to interstitial widening. There was no evidence of fibrosis or granuloma formation in this group (Figure 3). In Group 4, anthracotic pigment deposition was seen to co-exist with granulomatous inflammation in 10/32 cases (31.3%). The granulomas were non-caseating epithelioid cell type and showed the presence of Langhans' giant cells (Figure 4). Ziehl-Neelsen stain for acid-fast bacilli (AFB) was performed on tissue, AFB culture was done to confirm the diagnosis of TB in this group and

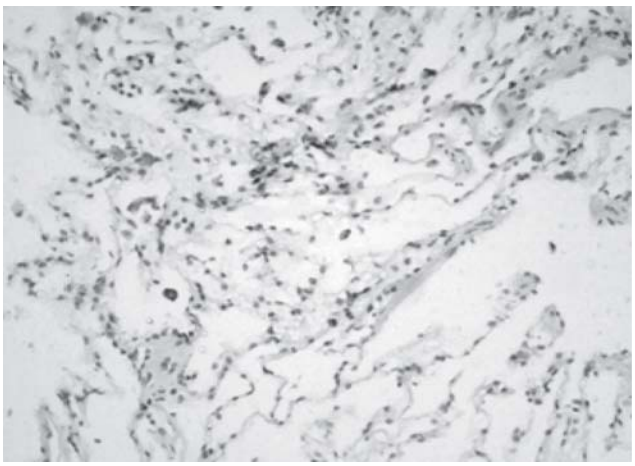

Figure 1. Photomicrograph of transbronchial lung biopsy showing normal lung parenchyma in controls (Group 1) (Haematoxylin and Eosin × 200).

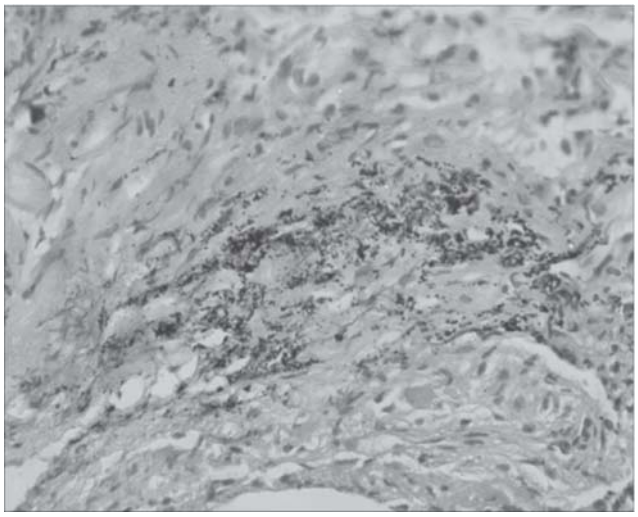

Figure 2. Photomicrograph of transbronchial lung biopsy showing anthracotic pigment deposition with fibrotic parenchymal reaction in Group 2 cases (Haematoxylin and Eosin × 100).

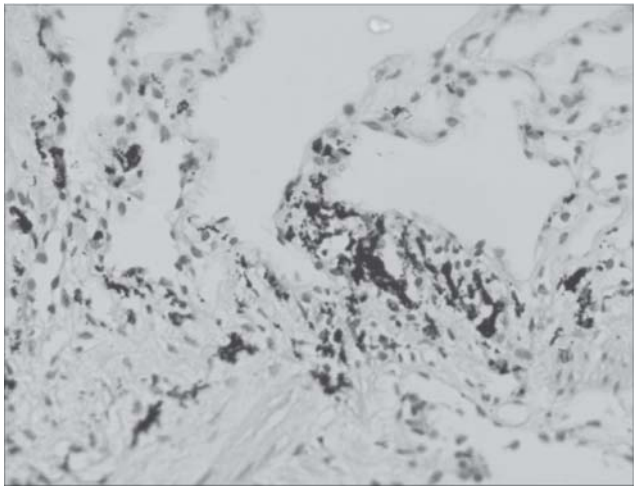

Figure 3. Photomicrograph of transbronchial lung biopsy showing anthracotic pigment deposition with interstitial inflammatory parenchymal reaction in Group 3 cases. There is no evidence of fibrosis (Haematoxylin and Eosin × 100).

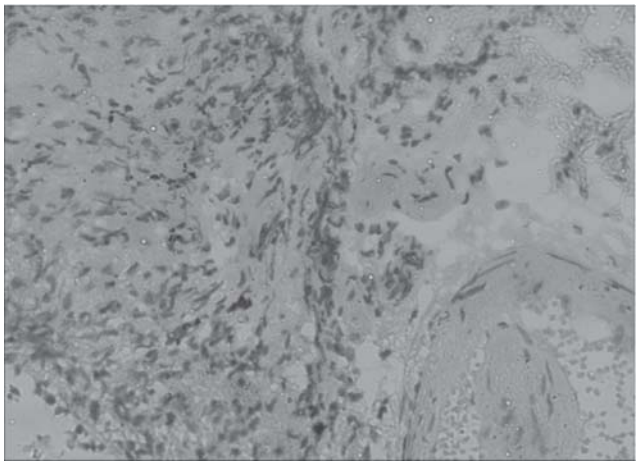

Figure 4. Photomicrograph of transbronchial lung biopsy showing perivascular epithelioid cell granuloma with anthracotic pigment deposition in Group 4 cases (Haematoxylin and Eosin × 200).

patients were started on anti-TB therapy.

Co-existent granulomatous inflammation was identified to be TB in 8/10 and sarcoidosis in 1/10 cases. However, the exact cause of their co-existence could not be identified. These have previously been suggested to be caused by superimposed infection on anthracofibrosis or an immune deficiency state produced by anthracofibrosis.[8]

Discussion

Bronchial anthracosis is defined as the deposition of multiple black-coloured carbon anthracotic pigmentation on large airway mucosa with or without airway narrowing or obliteration.[6] Individuals developed physical and radiologic abnormalities of the lung which were similar to chronic obstructive or fibrotic lung disease and can lead to a diagnostic dilemma. The diagnosis needs to be based on the recognition of the classic pathology seen on TBLB and its correlation with clinical and radiological features. Previously, anthracosis has been reported in 3% to 21% of all patients undergoing bronchoscopy.[8] This is similar to the present study in which the evidence of anthracotic pigment deposition was seen in the parenchyma of 32/384 (8.3%) TBLBs analysed.

In our series most of the patients presented with cough, dyspnoea and haemoptysis and underwent bronchoscopy to rule out underlying diffuse parenchymal lung diseases. On identification of anthracotic pigment in the TBLB, a thorough investigation was done for associated active or old TB infection, smoking (cigarette/*bidi*), air pollution, biomass exposure (*chullah*) and coal workers pneumoconiosis in all these cases as per reported literature.[10] *To the best of our knowledge, this is the first study from India where deposition of anthracotic pigment was correlated with the clinical and histopathological features.*

The link between TB and anthracosis has previously been proposed.[11] In this TB hypothesis, the bronchial infiltration by the adjacent TB lymphadenopathy is considered to continuously fistulise the wall of the bronchus with caseous granulomatous inflammation and later lead to local fibrosis,[9] as well as cause dark pigmentations.[6] However, whether *Mycobacterium tuberculosis* is one of the main causes in the development of bronchial anthracosis or that people with bronchial anthracosis are more susceptible to pulmonary TB are still unclear and further studies are required. Earlier studies from Asia had reported high frequency of TB (nearly 60%) in anthracofibrosis,[4] but recent studies from Korea and Iran have reported a decline in its frequency (20%-34%).[12-14] The frequency of TB in anthracofibrosis is even lower in developed countries.[15] In the western world, an uncommon

association of pulmonary TB with bronchial anthracofibrosis has been reported in immigrants from Indian subcontinent.[16]

Co-existent biomass/indoor air pollution/*chullah* smoke or occupational dust exposure were additional aetiological factors observed in our patients. This is similar to previously documented reports of anthracosis in patients with chronic exposure to biomass fuel smoke[17] and this entity has been termed as "hut lung".[7] Chronic wood smoke exposure has been linked with the deposition of anthracotic pigment in the macrophages, in epithelial cells, in the submucosal lung parenchyma and around the peripheral airways with anthracotic granule production at this site.[18] Interstitial inflammation and fibrosis[19] follows, eventually resulting in diffuse parenchymal lung diseases associated with anthracosis. Indoor air pollution accounts for nearly 1.5 to 2 million deaths per year worldwide, due to chronic obstructive pulmonary disease (COPD) and lung cancer, especially in children younger than five years and in women.[20] The inflammatory and fibrotic lung interstitial responses due to the various components of biomass smoke including anthracotic deposits have also been called as "fly ash lung".[21] Several hypotheses have been postulated for the development of indoor air pollution from solid fuels and progression of fibrosis. The indoor exposure to the incomplete combustion smoke of biomass fuels in places with limited ventilation, results in endobronchial deposit of anthracite with an inflammatory response that eventually leads to the development of chronic obstructive or fibrotic lung disease.[22]

Majority of patients in our study were cigarette/*bidi* smokers. However, since carbon is an inert element and known to elicit little or no fibrosis, it was deliberated that the retained anthracotic particles alone were responsible to induce bronchial stenosis in patients of bronchial anthracofibrosis. The additional pathology caused by concurrent mixed dust exposure, smoking, TB, other repeated exposures in these patients caused persistent inflammation and played a significant role in the pathogenesis and progression to diffuse parenchymal lung diseases. The mixed causal hypothesis that combines TB and inhalation hypotheses[9] is, therefore, advocated. This hypothesis suggests that exposure to biomass combustion smoke and or cigarette smoke leads to reduced mucociliary clearance and reduces the activity of the alveolar macrophages as well as the cellular immune response.[14,23,24] These tissue responses favour the development of TB and the frequent co-existence of both the entities. In the present study, silica crystals were identified by polarising microscopy of TBLBs. Their role in pathogenesis of lung parenchymal changes has been previously studied in humans and experimental animals, in whom silica potentiation of *Mycobacterium tuberculosis* growth occurs in macrophages[25] and aggravation of TB infections by silicosis has been demonstrated. Patients exposed heavily to air pollutants, cigarette smoke and biomass fuel smoke have carbon and silica deposit in their lymph nodes.[23] When these lymph nodes are infected with *Mycobacterium tuberculosis*, these rupture into the adjoining tracheobronchial tree, leading to black pigmentation and subsequently inflammation and fibrosis.

Conclusions

In the present study we demonstrate that anthracotic pigmentation in TBLB is not an innocent bystander. A categorical analysis of the TBLB pathology, an accurate identification of the pigment deposition and correlation with the underlying pathology in the limited tissue sample available needs to be done in order to reach a definitive diagnosis.

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