

Diagnosis of pulmonary disease in human immunodeficiency virus infection: role of transbronchial biopsy and bronchoalveolar lavage

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ABSTRACT The value of transbronchial biopsy and bronchoalveolar lavage was assessed in the diagnosis of pulmonary disease in patients infected with the human immunodeficiency virus (HIV). Seventy four transbronchial biopsy and 66 bronchoalveolar lavage specimens (60 paired specimens) from 80 examinations in 64 patients were reviewed. *Pneumocystis carinii* was the most common pathogen isolated (43 patients). Bronchoalveolar lavage was superior to transbronchial biopsy for the diagnosis of this pathogen, the sensitivities being 90% and 56%. Cytomegalovirus was identified three times by lavage and once by transbronchial biopsy. Neither method detected Kaposi's sarcoma in the one patient shown to have it by open lung biopsy. The complication rate in a concurrent study of bronchoscopy with transbronchial biopsy in 74 consecutive HIV positive patients was 22%. This study does not support the use of transbronchial biopsy in these patients.

Introduction

Lower respiratory tract infection and pulmonary Kaposi's sarcoma are features of the acquired immune deficiency syndrome (AIDS), and are responsible for much of the morbidity and mortality from this condition. Bronchoscopy is commonly performed in the investigation of such patients. Bronchoalveolar lavage and transbronchial biopsy are often performed at bronchoscopy in the belief that this gives the best chance of establishing the cause or causes of the pulmonary infiltrate. The value of the two procedures in management and their contribution to morbidity need to be assessed so that efficient investigation is carried out. This is important because of limited resources and the rising incidence of AIDS—the number of cases of AIDS reported in the United Kingdom to the end of August 1988 was 1730, the number of new cases doubling every 11 months.¹

Methods

Seventy four transbronchial biopsy specimens were reviewed histologically and 66 bronchoalveolar lavage

specimens cytologically. The specimens were obtained from 80 bronchoscopic examinations performed at the Middlesex Hospital, London, from August 1986 to October 1988 on patients with human immunodeficiency virus (HIV) antibodies and with clinical evidence of lower respiratory tract disease. Only 60 paired transbronchial biopsy and lavage specimens, however, were available for comparison. Four open lung biopsy specimens were also reviewed. The 80 examinations had been carried out on 64 patients; some had repeated episodes of respiratory disease and investigations. The single female patient had a history of intravenous drug abuse three years previously. The remaining patients were male homosexuals.

Transbronchial biopsy specimens were taken without fluoroscopic control, four specimens being obtained on each occasion. For lavage, 180 ml of warmed buffered saline* was instilled in 3 × 60 ml aliquots, usually into subsegments of the right lower lobe. A return of 45–70% of the fluid instilled was achieved. Lavage fluid was divided for cytological and microbiological studies. Biopsy specimens were formalin fixed and prepared in routine fashion. They were examined after being stained with haematoxylin and eosin, periodic acid-Schiff, Ziehl-Neelsen, and Grocott's silver methenamine.

Lavage specimens for cytological examination were
*Buffering: 0.75 ml of 8.4% NaHCO₃ in 1 litre of normal saline (0.9%).

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Table 1 Diagnoses from bronchoscopy and open lung biopsy specimens in 64 patients with human immunodeficiency virus antibodies

Diagnosis	Bronchoscopy (n = 80)		Open lung biopsy (n = 4)
	Transbronchial biopsy (n = 74)	Lavage (n = 66)	
Pneumocystis	30	34	
Cytomegalovirus	1	3	
Mycobacteria	1*		1*
Kaposi's sarcoma			1
Actinomycosis		1	

*Granuloma.

received in sealed centrifuge tubes and processed in a room designated for AIDS related pathology, the safety procedures for category B1 organisms² being followed. The fluid was centrifuged (1500 rev/min for five minutes). Deposit was spread on glass slides with disposable forceps. Slides were air dried and then alcohol fixed and left in alcohol for 15–20 minutes for decontamination before staining by the Papanicolaou and May-Grünwald-Giemsa methods. Most specimens were also stained with silver methenamine and Ziehl-Neelsen stains.

In a separate but concurrent study³ (including many of the same patients) we examined the complication rate in 93 consecutive HIV antibody positive patients undergoing bronchoscopy with transbronchial biopsy and lavage (74) or lavage alone (19) for respiratory problems. All patients had a normal platelet count and normal results in clotting studies.

Results

A specific pathological diagnosis was made on 50 occasions from 84 diagnostic procedures (bronchoscopy plus open lung biopsy). Pneumonia due to *Pneumocystis carinii* was diagnosed on 43 out of 50 occasions. Cytomegalovirus infection was diagnosed on three occasions, and mycobacterial infection twice; actinomycosis and Kaposi's sarcoma were each diagnosed once. All pneumocystis and cytomegalovirus infections were diagnosed by bronchoscopic examination. Mycobacterial infection and Kaposi's sarcoma were diagnosed by open lung biopsy (table 1).

PNEUMOCYSTIS PNEUMONIA

The relative sensitivities of transbronchial biopsy and cytological examination of lavage specimens were compared in 60 cases in which paired specimens had been obtained at the same bronchoscopic examination (table 2). Pneumocystis pneumonia was diagnosed in 39 patients; lavage gave a positive result in 32 cases, whereas transbronchial biopsy was positive in only 22 cases (56%). In only one case was *P carinii* diagnosed from a transbronchial biopsy specimen when

cytological examination had failed to show it. These figures include 16 transbronchial biopsy specimens considered technically "unsatisfactory" because of the small quantity of alveolar tissue obtained; *P carinii* was found in two of these specimens. A further three cases of pneumocystis infection were diagnosed by microbiological examination of lavage fluid. In three patients in whom both transbronchial biopsy and lavage had given negative results a diagnosis of pneumocystis infection was made subsequently—on the grounds of clinical suspicion and response to treatment in two and at necropsy in one. Overall, the sensitivity was 90% for lavage and 56% for transbronchial biopsy.

CYTOMEGALOVIRUS INFECTION

Cytomegalovirus inclusion bodies were detected in three lavage specimens, two being obtained from the same patient before and after treatment. In both cases the transbronchial biopsy specimen had been reported as negative, but serial sectioning and review after the cytological results were known led to the detection of cells with characteristic inclusion bodies. In the second case cytomegalovirus inclusion bodies were found in both the biopsy specimen and the lavage fluid. In none of the biopsy specimens was any appreciable "pneumonia" present. The patients' symptoms were related to cerebral and ocular cytomegalovirus infection. No concurrent infection with cytomegalovirus and pneumocystis was found.

MYCOBACTERIAL INFECTION

Careful examination of specimens failed to detect acid fast bacilli. One transbronchial biopsy specimen contained a poorly formed granuloma, possibly mycobacterial in origin, and in one open biopsy specimen a caseating granuloma was present, presumed to be mycobacterial; culture of fluid and lung tissue from both these patients was negative.

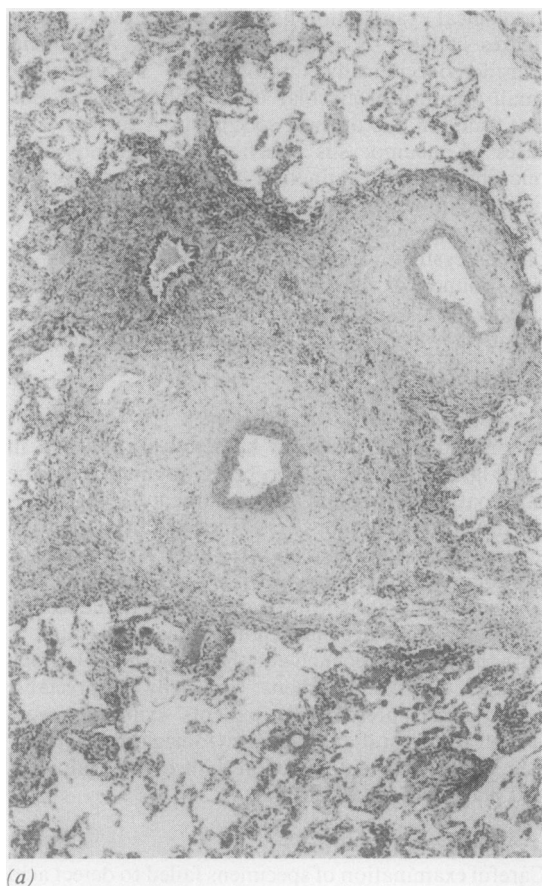
ACTINOMYCOSIS

Actinomycosis was identified in one lavage specimen but it was not seen in the corresponding transbronchial biopsy specimen.

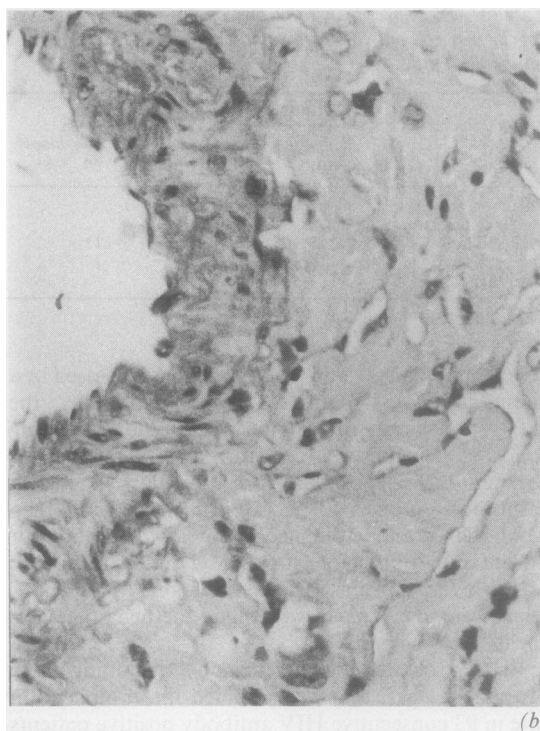
Table 2 Diagnosis of *Pneumocystis carinii* pneumonia from 60 paired transbronchial biopsy (TBB) and bronchoalveolar lavage fluid (BAL) specimens

	TBB (histology)	BAL (cytology)	Combined TBB and BAL
Positive	22	32	33
Negative*	38	28	27
Total	60	60	60

*Six patients with negative results from transbronchial biopsy and bronchoalveolar lavage had pneumocystis pneumonia diagnosed by other means (3), by the response to treatment (2), or at necropsy (1).



(a)



(b)

Pulmonary Kaposi's sarcoma: (a) low power view showing expansion of perivascular and peribronchiolar connective tissue by Kaposi's tissue; (b) high power view showing the characteristic angular endothelial lined spaces of Kaposi's sarcoma separated by hyalinised collagen, an unusual feature.

KAPOSI'S SARCOMA

Extensive Kaposi's sarcoma was found in one patient from open lung biopsy (figure). The patient, who had cutaneous Kaposi's lesions, had had two transbronchial biopsies with negative results. Immunostaining for the endothelial markers factor VIII antigen and *Ulex europaeus* lectin binding was unhelpful. Open lung biopsy showed the Kaposi's sarcoma to be infiltrating interlobular septa, pleura, and peribronchial and perivascular tissues (figure, *a*). An unusual feature was the dense collagenous sclerosis between the ectatic vascular spaces, which were similar in other respects to those of the "patch" stage of cutaneous Kaposi's sarcoma (figure, *b*). In only a few areas were "nodular" lesions found. The sclerosis may have been secondary to hilar lymphatic obstruction by Kaposi's sarcoma.

COMPLICATIONS

The complication rate for the 74 bronchoscopies with concurrent transbronchial biopsy was 22%. This included 15 episodes of pneumothorax, eight of which

required a chest drain, and two cases of profuse haemorrhage, one of which was fatal. Pneumothorax occurred in 14 (27%) of 52 patients with pneumocystis infection compared with one in the 22 without pneumocystis infection. The only complication in the 19 patients having lavage alone was bronchial wall oozing in two cases.

Discussion

Bronchoscopy is performed in HIV infected patients with clinical evidence of lower respiratory tract disease to provide bronchoalveolar lavage fluid for microbiological and cytological investigations, and to provide transbronchial biopsy specimens for histological examination. The airways may be examined at the same time for evidence of bronchial Kaposi's sarcoma. In some centres transbronchial biopsy specimens are used to make touch preparations for cytological assessment as well as for histological examination. The aim is to detect opportunist infection and pulmonary Kaposi's sarcoma so that specific

treatment may be given or, in patients in whom a pathological diagnosis has already been established, to determine the response to treatment. In addition, detecting these conditions to establish the diagnosis of AIDS in a patient known to have HIV antibodies may be important. Non-opportunist infections, also common in these patients,⁴⁻⁶ may be diagnosed in this way, as may incidental inflammatory and neoplastic processes.

Pneumocystis carinii was the most common organism identified, as in other series based on biopsy or cytology. In two postmortem studies,^{7,8} however, cytomegalovirus was the most commonly identified pathogen.

We found bronchoalveolar lavage superior to transbronchial biopsy for detecting *P. carinii*. Other series have found a similar high sensitivity for bronchoalveolar lavage⁹⁻¹¹—95% in 20 cases in which lavage findings were compared with necropsy.¹¹ Broaddus *et al.*¹² however, reported a high sensitivity for both transbronchial biopsy and bronchoalveolar lavage (93% and 89%) in 117 paired specimens; whereas a collective review of 141 cases recorded a sensitivity of only 79% for the cytological detection of *P. carinii*.¹³ Variations in technique, such as bronchial washing rather than bronchoalveolar lavage, may account for the reported lower sensitivity of cytology. A sensitivity of only 79% for cytology and 85% for transbronchial biopsy in paired specimens has also been reported, but the figure for cytology appears to include touch preparations from transbronchial biopsy as well as bronchoalveolar lavage specimens and this may explain the lower yield.¹⁴ It is not surprising that bronchoalveolar lavage is more effective than transbronchial biopsy for detecting pneumocystis infection, as the organisms lie almost entirely within the alveolar space and the alveolar content of at least 50 cm³ of lung is sampled during lavage, compared with less than 1 cm³ by transbronchial biopsy.

In our two patients with cytomegalovirus infection bronchoalveolar lavage was superior to transbronchial biopsy in identifying cytomegalovirus inclusion bodies. Culture is superior to both^{15,16} but takes longer. The detection of early antigen by the fluorescent foci test (DEAFF) in bronchoalveolar lavage fluid rapidly identifies cytomegalovirus shedding but gives no indication of cytomegalovirus cytopathology. The meaning of a positive DEAFF test in these patients is still being evaluated. The incidence of cytomegalovirus infection in postmortem studies^{7,8} is very much higher than in antemortem series. Most cells showing the pathological features of cytomegalovirus infection appear to lie free within the alveolar space and bronchoalveolar lavage would be expected to give a higher yield than transbronchial biopsy. The importance of finding pulmonary cells infected with cyto-

megalovirus is that disease in other tissues less accessible to biopsy (brain, eye) may then be attributed to cytomegalovirus. Pulmonary infection alone is rarely an indication for treatment.

Acid fast bacilli were not detected by bronchoalveolar lavage or transbronchial biopsy in this study. Infection with *Mycobacterium tuberculosis* is common in intravenous drug users with AIDS,¹⁷ but the lungs are less often affected than other organs.¹⁷⁻¹⁹

Kaposi's sarcoma was not identified by the two transbronchial biopsies from a patient in whom it was diagnosed by open lung biopsy. Bronchial Kaposi's sarcoma is easily identified bronchoscopically and in bronchial biopsy specimens,²⁰ but parenchymal Kaposi's sarcoma is difficult to diagnose by transbronchial biopsy,²¹ though haemosiderin deposits and haemorrhage may give grounds for suspicion. Open lung biopsy is usually necessary to make the diagnosis in life,²² but it may also fail to show tumour in life.²³⁻²⁵ Kaposi's sarcoma has been diagnosed by transbronchial biopsy.^{14,26}

Transbronchial biopsy did not contribute appreciably to the diagnosis of lower respiratory tract disease in our HIV infected patients. Histological examination provides information about the nature of the cellular infiltrate and its extent in the small piece of lung parenchyma sampled. It might suggest a diagnosis such as non-specific interstitial pneumonitis,²⁷ but the therapeutic implications of this information are negligible. Among 10 patients with a diagnosis of non-specific interstitial pneumonitis from transbronchial biopsy, a specific diagnosis was found in seven at open lung biopsy.²² We confirmed that bronchoalveolar lavage was useful in diagnosing pneumocystis pneumonia. The cytological technique requires care but no particular expertise or experience. Neither bronchoalveolar lavage nor transbronchial biopsy was very successful in identifying cytomegalovirus infection.

Our complication rate of 22% in patients having transbronchial biopsy was due to the high incidence of pneumothorax, mainly in patients with pneumocystis infection. This compares with reported pneumothorax rates of 9–19% for transbronchial biopsy,^{12,28-30} which is greater than for bronchoscopy alone.³¹ Fluoroscopic control made no difference to the complication rate of transbronchial biopsy in a series of 250 bronchoscopies in patients with AIDS.³² Transbronchial biopsy in these patients is difficult to justify because the small contribution made to diagnosis was offset by the complications associated with it.

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