Bronchoalveolar Lavage in Occupational Lung Diseases

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ABSTRACT

Occupational lung diseases (OLDs) are related to the exposure and inhalation of organic, inorganic, and synthetic particles, fumes, gases, or infectious agents. From the long list of OLDs this article focuses the discussion on bronchoalveolar lavage (BAL) in parenchymal immunoinflammatory conditions, such as hypersensitivity pneumonitis (HP) and pneumoconiosis.

Several antigens may cause HP, including products of plant or animal origin, aerosolized microorganisms, and organic chemicals. BAL is used not only to assess the pathogenesis of these diseases but also to identify the typical pattern of intense lymphocytic alveolitis, usually with a CD4:CD8 ratio below normal and frequently with the presence of mast cells, plasma cells, and foamy macrophages.

Pneumoconioses are chronic interstitial lung diseases caused by the inhalation of mineral and metallic inorganic particles/dusts in an occupational setting, showing a decreasing prevalence in recent years.

BAL is a useful tool not only to express the complex pathogenic mechanisms of these entities but also in excluding other diagnoses and causes of alveolitis, and to document specific exposures, such as the identification of asbestos bodies (ABs) in asbestosis or the proliferative response of BAL lymphocytes to beryllium in chronic beryllium disease (CBD).

KEYWORDS: Bronchoalveolar lavage, occupational lung diseases, hypersensitivity pneumonitis, pneumoconiosis

The risks of injury and lung disease have been recognized in mining since the 16th century and were elegantly described at that time by Agrícola.

Currently we are aware that there is an ongoing increase of occupational lung diseases (OLDs) in several developed or developing countries, and that, in Western countries, there is a shift in these diseases from pneumoconiosis to occupational asthma, the current most common work-related respiratory event.[1]

This is closely related to industrial development and the improvement of new biotechnologies, with changes not only in agriculture and basic industries, like mining and metallurgy, reflected in fewer risk populations, better hygiene measures, and less harmful exposure, but also in the widespread use of enzymes, leading to a higher degree of sensitized workers, expressed in an increasing number of allergic occupational diseases.[2]

However, in most countries, there are still inadequate data to determine the number of injured workers or those at risk, and the association between exposure to a potentially harmful agent and lung disease is underestimated.

Nevertheless it seems that OLDs are, in terms of frequency, severity, and preventability, the most common work-related illnesses in the United States.[3]

In this context, there were ~22.000 newly reported occupational respiratory conditions in the United States during the year 2002, and during the same period, it is estimated that nonfatal OLDs were under progression in overall 2.5 per 10.000 full-time workers.

OLD may develop through exposure and inhalation of organic, inorganic, and synthetic particles, fumes, gases, or infectious agents, under an isolated or multifarious context, in the workplace or even due to a particular hobby. It is essential to confirm or rule out an occupational etiology, not only through a detailed occupational history but also by searching for confounding or related factors, like smoking habits, exposure to other pollutants, and certain hobbies.[5]

These exposures can lead to several conditions, like airways diseases (asthma, occupational bronchitis, and acute inhalation disorders), infectious and malignant conditions, hypersensitivity pneumonitis (HP), and pneumoconiosis, expressed in a long list of clinical entities.

The discussion here focuses on bronchoalveolar lavage (BAL) in OLD parenchymal immunoinflammatory conditions, such as HP and pneumoconiosis.

HYPERSENSITIVITY PNEUMONITIS

HP, also known as extrinsic allergic alveolitis, is an inflammatory and granulomatous interstitial lung disease caused by intense and/or repeated inhalation of organic dusts, with further sensitization.

Although HP was recognized by Ramazinni, in 1713, through the report of wheat reapers with this clinical pattern,[6] the first description of the disease was made by Campbell in 1932, in farmers exposed to moldy hay, with so-called farmer's lung.[7]

Today, there are more than 200 different antigens underlying HP, consisting of products of plant or animal origin, aerosolized microorganisms, and organic chemicals that must have a diameter below 3 to 5 μm to be inhaled and reach the lung periphery.[8]

The most frequent antigens are listed in Table 1 and are related to a variety of distinct occupations in different places of the world, from plaster workers exposed to esparto dust in the southeast Iberian

Table 1 Main Causes of Hypersensitivity Pneumonitis

Antigen/Agent	Source	Disease
Thermophilic actinomycetes— *Micropolyspora faeni*, *Thermoactinomyces vulgaris*, *T. saccharii, T. viridis, T. candidus*	Moldy hay, moldy compost, moldy plant materials, moldy sugarcane residue	Farmer's lung Ventilation pneumonitis Bagassosis Mushroom worker's disease
Avian proteins	Bird droppings/feathers/serum	Bird-breeder's lung, bird-fancier's lung, pigeon-breeder's lung
Rat urinary proteins	Rat urine or serum	Laboratory worker's hypersensitivity pneumonitis
Trichosporon cutaneum	Mold in Japanese homes	Summer-type hypersensitivity pneumonitis
Isocyanates	Paints, plastics	Isocyanates hypersensitivity pneumonitis
Anhydrides	Plastics	Epoxy-worker's lung
Penicillium glabrum (frequentans)	Moldy cork	Suberosis
Penicillium casei	Cheese mold	Cheese-washer's lung
Aspergillus clavatus	Moldy grains	Malt-worker's lung
Cryptostroma corticale	Moldy maple bark	Maple bark-stripper's disease
Aureobasidium pullulans	Moldy sequoia dust	Sequoiosis
Amoeba—*Naegleria gruberi*, *Acanthaemoeba castellani*	Contaminated heating, ventilation, and air conditioning systems and humidifiers	Ventilation pneumonitis
Pyrethrum	Insecticides	Insecticide lung
Lycoperdon species	Puffballs	Lycoperdonosis
Bacillus subtilis proteins	Contaminated wood dust	Woodworker's
Bacillus subtilis enzymes	Detergent enzymes	Detergent-worker's lung
Mycobacterium avium-intracellulare	Contaminated water	Hot tub lung
Aureobasidium	Contaminated water	Sauna-taker's disease

Peninsula[9] to people working in the mushroom crops of Bunashimeji in Japan.[10]

The prevalence of HP is related to several factors, such as the type, intensity, and duration of exposure; the antigen concentration and solubility; particle size; and host susceptibility. It is estimated that, in the population exposed to recognized inciting agents, the prevalence is between 5 and 15%,[11] although a larger percentage of individuals may show sensitization, even without lung disease, such as potentially in ∼40% of pigeon breeders.[12]

The antigens, once within the alveolar structures and dependent on the yet described variables, may induce cellular and humoral immune reactions, such as immune complex and cell-mediated immunological disorders. The knowledge acquired about these mechanisms is mostly derived from BAL studies, which are under a clinical or experimental scope.

In this context, BAL may be used to assess the pathogenic pathway of HP, namely, through the evaluation of cytokine levels. Wolff et al[13] described how fibronectin and transforming growth factor-β are significantly increased in HP patients compared with eosinophilic pneumonia, cryptogenic fibrosing alveolitis, and sarcoidosis individuals, possibly related to a higher intensity of inflammation in HP, and interferon-γ is significantly decreased in HP compared with sarcoidosis, probably caused by the Th2 vs Th1 bias in both diseases.

Yet the most typical pattern of BAL in HP is the presence of an intense alveolitis, showing a three- to fivefold increase in cell counts, with a clear predominance of lymphocytes, frequently achieving percentages higher than 50%,[8,14] usually with a CD4:CD8 ratio below normal, in which this lymphocytic alveolitis is a major criterion for the diagnosis of HP in the absence of pathological confirmation.[15]

Set against other interstitial lung diseases (ILDs), the presence of mast cells is more frequent in HP; also plasma cells are frequent in HP and in a small number of other clinical entities such as organizing pneumonia, chronic eosinophilic pneumonia, and aspiration pneumonia.[14] Macrophages often exhibit particular morphological changes, namely, a foamy cytoplasm.

Nonetheless, these BAL findings are dependent on the timing of the last antigen exposure; neutrophils are elevated within 48 hours after acute exposure and values return to normal within a week. On the other hand, lymphocyte levels remain increased a long time after removal of the inciting antigen, sometimes years, but in lower percentages compared with patients still exposed. When progression leads to fibrosis, neutrophils may again be increasingly present in HP and CD4:CD8 ratio may show mild elevation.[8,11]

The different lymphocytic phenotyping was reported by Ando et al[16] in 271 patients with summer-type HP (CD4:CD8 = 0.6 ± 0.1) compared with 22 farmer's lung HP (4.4 ± 0.7). This different CD4:CD8 ratio, with important implications in the immunopathogenesis of HP, may depend on several factors, such as (1) the type and dose of inhaled antigen (being intermittent exposures to *Micropolyspora faeni* eventually related with the high levels found in farmer's lung and more continuous exposures to low concentrations of *Trichosporum cutaneum* reflected in the diminished numbers of summer-type HP); (2) the characteristics of granuloma (smaller and immature in summer-type HP); (3) the staging of disease, as previously stressed, and the therapy received; (4) the existence of other inhalants (eventually present in farmer's lung patients); and (5) smoking habits (higher CD4:CD8 ratio in farmer's lung smokers vs nonsmokers). Interestingly, more HP cases occur in nonsmokers, which is likely due to the immunosuppressive effects of tobacco, reflected in a decrease in the production of specific antibodies to the causative antigens.[8]

Concerning neutrophils, Pardo et al[17] showed a positive correlation between the percentage of lung tissue polymorphonuclear leukocytes (PMN) and the percentage of lung fibrosis in 15 HP patients, but there was no correlation between BAL and lung neutrophils.

Some investigators suggested correlations between the number of neutrophils in BAL and either the severity or the response to therapy or both in idiopathic pulmonary fibrosis (IPF)[18] or the activity and prognosis of sarcoidosis[19] or even with the fibrotic evolution in HP.[17,20] However, these findings were not confirmed by other studies.[14] The importance of BAL PMN as a marker of activity of disease or disease evolution requires further investigation.

However, the most important diagnostic information obtained from BAL cellularity is that normal values rule out HP, making BAL a sensitive tool in the differential diagnosis with other ILDs.[5,8,11,14,15,21] In this context, Drent et al developed a software program, using patients (gender and age) and BAL (percentage of recovery, total cellularity, and quantification of macrophagic, lymphocytic, and neutrophilic populations) data. A logistic regression mathematical analysis is applied to these data and used as a method of high positive predictive value to differentiate the most prevalent ILD disorders: HP, sarcoidosis, and idiopathic pulmonary fibrosis.[22,23]

Nevertheless, there may be considerable overlap with BAL data when considering HP and other entities, namely, nonspecific interstitial pneumonitis (NSIP) and the newly described airway-centered interstitial fibrosis (ACIF) pattern.[24,25]

Moreover, BAL does not differentiate between healthy, sensitized individuals—the so-called subclinical alveolitis (SA)—and HP patients, only allowing the exclusion of nonsensitized individuals.

Table 2 Bronchoalveolar Lavage Data on Hypersensitivity Pneumonitis—Pulmonology Research Centre

Total Cells $\times 10^4$	Macrophages %	Lymphocytes %	Neutrophils %	Eosinophils %	CD4:CD8
33.7 ± 16.3	59 ± 29.6	35 ± 28.9	4 ± 5	2 ± 2.9	0.9 ± 0.5

Numerous studies have been performed in this field, in the Research Centre of Pulmonology–University of Coimbra (CPUC), on SA in several occupational and nonoccupational diseases, as in pigeon breeders, patients treated with amiodarone, workers exposed to asbestos fibers and crystalline silica, and in cutaneous sarcoidosis, connective tissue diseases, and patients with human immunodeficiency virus (HIV) infection.[26–31]

Although uncovering these BAL changes in individuals without respiratory symptoms, with a normal chest x-ray and normal lung function, could be of obvious interest for the clarification of pathogenic mechanisms and for supporting the cessation of exposure to the likely related agents (antigens, medications, or others), the significance of SA is still discussed, as a local expression of systemic diseases or as an isolated pulmonary involvement. The prognostic implication remains obscure because additional cellular and mediator data and more comprehensive longitudinal studies are needed.[32,33]

In a retrospective analysis of the last 20 consecutive patients with HP with different causes submitted to BAL in the CPUC, the authors confirmed the existence of a lymphocytic alveolitis with diminished CD4:CD8 values (Table 2).

From this retrospective study, the authors single out a patient with one of the most prevalent HPs in Portugal; a 45-year-old nonsmoking businessman who was referred to the pulmonology outpatient clinic because of a 5-month history of persistent cough productive of sputum, exertional dyspnea, wheezing, and fatigue. He bred chickens, pigeons, turtle-doves, and canaries.

Chest x-ray revealed a bilateral reticulonodular pattern, and high-resolution computed tomography (HRCT) showed ground-glass attenuation (Fig. 1).

Lung function indicated a restrictive ventilatory defect with a forced vital capacity (FVC) of 60.07%, forced expiratory volume in 1 second (FEV1) of 62.6%, and FEV1/FVC of 86.7%, with severe reduction of diffusion capacity "transfer lung capacity (TLCO)" of 37.8%.

BAL findings included a significant increase in total cell count (74×10^4 cells/mL), with lymphocytic predominance (80% of total cells), showing also the presence of plasma cells, mast cells, and foamy macrophages (Figs. 2 and 3).

Laboratory findings included precipitating antibodies "immunoglobulin G (IgG)" for pigeon feathers, droppings, and serum and negative results for chickens, turtle-doves, and canaries.

This suggestive clinical picture of HP related to pigeon breeder's disease, also reinforced by the interstitial infiltrate of lymphocytes, plasma cells, neutrophils, and eosinophils found on histology obtained from a transbronchial biopsy (Fig. 4), strongly suggest allergen contact avoidance.

Indeed, after removal of the birds from the patient's home and a treatment with oral steroids (1 mg/kg/d of prednisone followed by gradual taper), there was significant clinical, radiological, and functional improvement, denoted by the absence of symptoms after 4 months and also disclosed in Fig. 5 and Table 3.

Portugal is the largest cork-producing country in the world, with more than 1000 factories, employing about 15,000 workers; consequently, suberosis is particularly prevalent. Suberosis together with bird fancier's disease (BFD), represent two of the most frequent HP etiologies in this country. Suberosis is caused by the repeated inhalation of *Penicillium glabrum* (formerly known as *Penicillium frequentans*) that contaminates
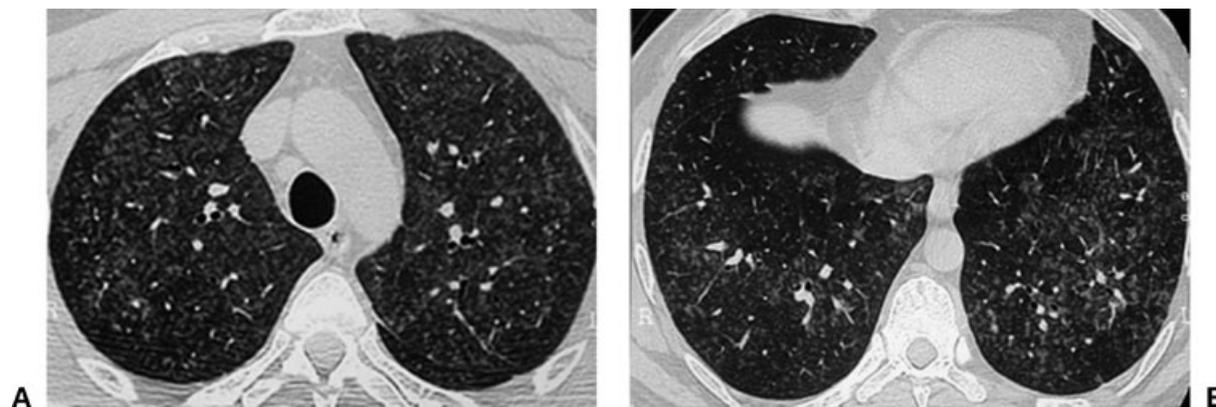
Figure 1 High-resolution computed tomography with ground-glass attenuation in (A) upper and (B) lower lobes.
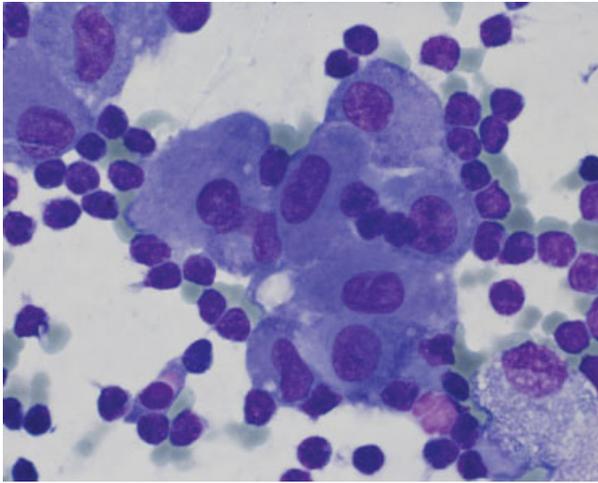
Figure 2 Lymphocytic alveolitis, activated macrophages (one showing foamy cytoplasmic alterations) and presence of a plasma cell. May-Grünwald Giemsa (MGG) × 100.
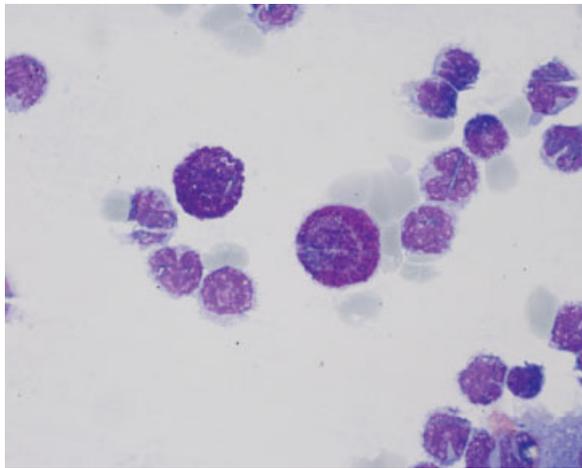

Figure 3 Presence of two mast cells in a slide with lymphocytic predominance. MGG × 100.
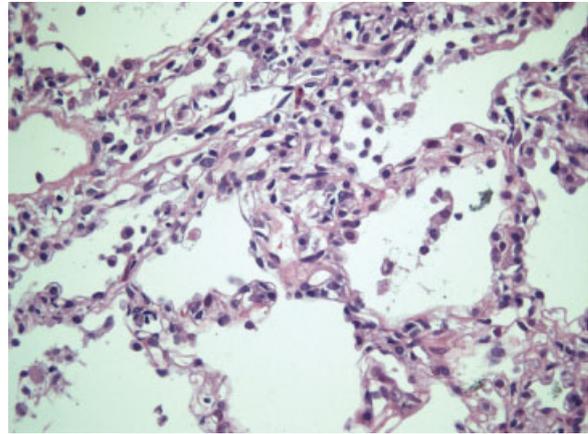

**Figure 4** Transbronchial biopsy showing interstitial inflammatory infiltrate.

cork during its industrial processing, and it is estimated that the prevalence of this disease among cork workers is between 9 and 19%.[34]

A large cohort of patients with these diseases (81 with suberosis and 32 with BFD) was recently described, allowing clinical, functional, radiological, and BAL patterns to be compared in both populations.[35]

Besides the findings of more acute forms and more severe restrictive defects in patients with BFD, BAL revealed more intense alveolitis in BFD (90×10^4 cells/mL vs 66×10^4 cells/mL in suberosis), with more lymphocytes, in absolute and also relative numbers (61.7% in BFD vs 58.8% in suberosis), and CD4:CD8 ratio was significantly more decreased in suberosis (0.47 vs 1.1 in BFD).

Comparing patients no longer exposed to those still exposed, the only difference was a diminished percentage of CD8 + cells in the case of suberosis individuals where the agent was withdrawn (49.3% vs 55.5%).

This study thus confirms the interest of BAL data by showing the existence of different immune and inflammatory responses, even in disorders with pathophysiological similarities.
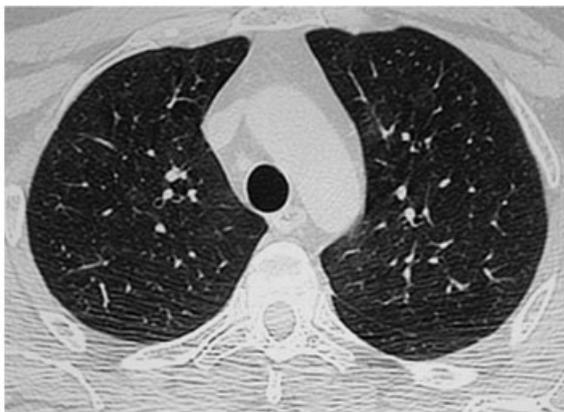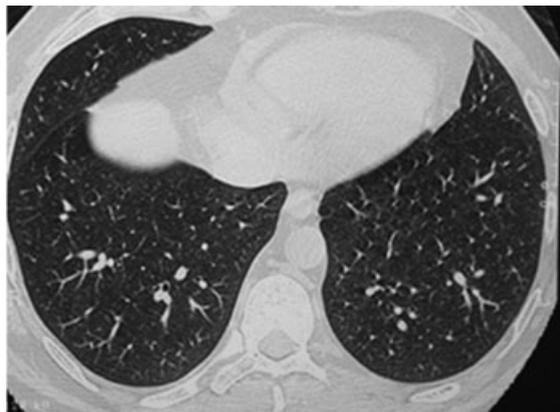
Figure 5 Normal high-resolution computed tomography—(A) upper and (B) lower lobes.

Table 3 Pulmonary Function Tests of Hypersensitivity Pneumonitis Patient: Follow-Up and Evaluation

Variable	Presentation (%)	4 Months Later (%)	10 Months Later (%)
FVC	60.07	98.41	106.8
FEV1	62.62	102.3	105.4
FEV1/TVC	86.72	86.42	81.94
TLC	67.01	85.34	77.40
TLCO (SB)	37.88	53.69	83.96

FEV1, forced expiratory volume in 1 second; FVC, forced vital capacity; SB, single breath; TLC, total lung capacity; TLCO, transfer lung capacity.

PNEUMOCONIOSES

Pneumoconioses are chronic ILDs caused by the inhalation of mineral and metallic inorganic particles/dusts in an occupational setting. The changing pattern of OLD is reflected, in developed countries, in a decrease in the prevalence of pneumoconioses and even in a better outcome of these entities, which is related to a tighter control and reduction of hazardous exposures, earlier diagnosis of the diseases, and the replacement of known aggressive industrial agents.[36] Nevertheless, there remain various occupations that may result in exposure to several causal agents of pneumoconioses,[37–40] summarized in Table 4.

The inhalation of agents such as crystalline silica may evoke lung reactive processes and injuries leading to pulmonary fibrosis. This depends on several factors, including the duration of exposure, the concentration of particles in the environment, the dimensions of the particles (particles reach the alveoli only if the particle diameter is less than 5 to 3 μm) and their morphological and physicochemical surface characteristics. Host factors such as individual worker susceptibility should also be considered.

The pathogenic mechanisms are complex, initially sustained by macrophagic and natural killer cell inter-vention, after opsonization of particles by surfactant and other substances. These changes may be expressed on BAL (and also on tissue samples) by alveolitis of lymphocytic predominance and sometimes also with a neutrophilic or mixed pattern. Usually, there is preferential activation of CD4 or CD8 lymphocytes and TH1/TH2 differentiation inducing progression to a granulomatous reaction or to fibrosis, with activation of interleukins-1,4,6,8,10,12, and 13, tumor necrosis factor (TNF)-α, matrix metalloprotease (MMP)-1, tissue inhibitor of metalloprotease (TIMP)-1, intercellular adhesion molecule (ICAM)-1, transforming growth factor (TGF)-β, epidermal growth factor (EGF), insulin growth factor (IGF), platelet-derived growth factor (PDGF), among others.[41,42]

In silicosis (the most prevalent chronic occupational disease in the world, resulting from inhalation of crystalline or free silica, usually in the form of quartz), it is possible to find a lymphocytic SA on initial exposure. However, alveolitis usually exhibits a mixed lymphocytic/granulocytic pattern, macrophages may be increased, and the CD4:CD8 ratio is decreased.

BAL is also important in excluding other diagnoses and causes of alveolitis, and to document specific dust exposures.

In this context the authors stress two entities where BAL findings are particularly useful, asbestosis and berylliosis.

Exposure to asbestos gives rise to pulmonary fibrosis (asbestosis), lung cancer, and pleural diseases, which may be benign (effusion, fibrosis, plaques) or malignant (mesothelioma).

Asbestos fibers are silicate fibers of different lengths and widths, from two main families: serpentines (chrysotile) and amphiboles (crocidolite, the most frequent, and also amosite, tremolite, anthophyllite, and actinolite). Chrysotile represents between 90 and 95%

Table 4 Main Causes of Pneumoconioses

Activities	Agent
Ceramic and pottery plants, sandblasting, mining industry, quarries, tunneling, foundry work, coal working and extraction, stone cutting, cement production, glazing, vitreous enameling	Free silica dust
Thermal and acoustic insulation, mining, shipyard activities, automotive industry, textile industry, production of friction materials	Asbestos
Mining industry, quarries, rubber and paper production industry, ceramic and pottery plants, cosmetics	Silicates and talc
Mining, smelting, grinding steel, polishing, arc welding	Iron
Diamond polishing, electroplating	Cobalt
China, fine earthenware, and pottery product manufacturing	Kaolin
Fluorescent and neon lamp industries, aircraft engines, guidance systems and brakes, x-ray tube windows, turbine reactor blades, dental castings and prostheses, electrical and electronic components, high-tech ceramics	Beryllium
Hard metal tools manufacturing	Hard metal
Explosives workers, blasters, aluminum powder workers	Aluminum

of the world production, namely, in Canada, Alaska, and the Russian Federation. Nevertheless, crocidolite has been identified as a more potent fibrogenic agent, so pulmonary fibrosis is more frequent in processing than in the mining industry. Moreover, intermittent exposures, for example, in shipyards, are related to a higher prevalence of pleural changes, whereas more continuous and cumulative exposures are related to parenchymal fibrosis.[36,42,43]

BAL is a useful tool in asbestosis, not only to assess the common moderate lymphocytic (and/or neutrophilic) alveolitis, higher in the initial phases (eventually as an SA), lower in more advanced disease, and usually associated with augmented CD4:CD8 ratio, but essentially to identify and quantify the asbestos fibers.

It is assumed that the number of asbestos bodies (ABs) found in BAL reflects concentration of these bodies in the lung.[44] More controversial is the choice of method to identify the asbestos and the significance of the quantitative counting of fibers. ABs can be seen directly on BAL slides but quantitative demonstration after filtration with the AB Millipore filtration system (Millipore, Billerica, MA) seems to be more sensitive, namely, when the level is low[14,45]; this filtration followed by light microscopy study of 20 mL of BAL is the method recommended by a working group from the European Respiratory Society (ERS).[46] Also, it is important to recognize and differentiate true ABs, with a thinner segmented rod, from the thicker pseudobody, with irregularities in its central fiber. On the other hand, ABs in BAL are frequently coated with proteic and iron material and are easily identified through light microscopy, whereas the uncoated fibers are only detected by electron microscopy.

In a recent paper, Romeo et al[47] compared the counting of ABs in BAL using a phase contrast microscope with the fiber concentration defined by transmission electron microscopy, in 193 asbestos-exposed workers and 84 nonexposed individuals. Electron microscopy revealed positive fibers in all the exposed individuals, whereas phase contrast microscopy detected ABs in only 75.1% of this population.

Nevertheless, BAL is a reliable, early, and sustained marker of asbestos exposure. The presence of ABs in BAL is associated with a higher prevalence of respiratory symptoms, reduced pulmonary function, and parenchymal changes.[45] The correlation between BAL and lung tissue has been presented either as one AB/mL of BAL reflecting 100 to 10.000 ABs/cm^3 in lung,[48] or ~1000 to 3000 ABs/gr of dry tissue.[49]

This concentration (more than one AB/mL of BAL or per 10^6 cells) is considered significant evidence for asbestos exposure[49–51] with the filtration technique, with sensitivity and specificity of 89%.[52] However, Costabel pointed out several more than 0.5 AB/mL of BAL as indicative of above-average exposure.[14] De

Vuyst et al[50] reported, in a large population, that 90% of asbestosis cases had $\geq$ 5 AB/mL.

However, very high levels of concentration in BAL are now less frequent because the use of asbestos is decreasing in most countries, namely, since the mid-1970s, after several regulations of exposure levels and prohibition of use were implemented. This was associated with changing patterns of exposure, from primary asbestos workers to those exposed to materials in buildings and industrial sites.[52]

In the case of the World Trade Center (WTC), ~5000 tons of asbestos were used for fireproofing in the construction of the towers. In this context, Rom et al[53] describe a clinical case of a New York City firefighter exposed to high concentrations of WTC dust for 2 weeks during the September 2001 crisis. Acute eosinophilia was noted as a rare consequence of acute high dust exposure. Besides fly ash and degraded glass, mineralogic analysis of BAL showed 305 uncoated fibers of asbestos per 10^6 cells using electron microscopy, with the absence of coated AB reflecting the recent acute exposure.

Negative BAL findings do not exclude increased AB parenchymal content, namely, when there is a mild AB count in lung tissue. Detection of ABs in the lung remains the "gold standard" for asbestos exposure. Nonetheless, the search for noninvasive procedures is mandatory, and even induced sputum (IS) is being used in this field.

However, IS seems to be much less sensitive than BAL and useful mainly in the setting of high levels of asbestos exposure.[54–56] To increase sensitivity, several samples should be obtained from each worker.[57]

Although Capellaro et al[58] found agreement of AB detection between BAL and IS in only 56.8% of their clinical cases, IS may be a complementary tool to BAL (or even substitutive in the case of contraindication to fiberoptic bronchoscopy), not only in asbestosis, but also in the evaluation of other pneumoconiosis patients (silica and hard metal workers). Fireman et al[59] found similar quantitative and qualitative results in the number and chemical analysis of particles obtained by BAL or IS. Thus IS could be useful as a biological monitoring method in periodic health examinations of healthy workers exposed to hazardous dusts and could be performed even in the workplace.[56]

The use of beryllium in industry began in the 1930s and is ongoing, although the number of workers exposed to this metal worldwide is not known.

The chronic beryllium disease (CBD), also known as berylliosis, is a granulomatous disease that has been reported in the United States, Canada, the United Kingdom, Germany, Poland, Israel, Japan, and South Korea. CBD affects ~1 to 3% of exposed workers and is clinically, radiologically, and histologically indistinguishable from sarcoidosis.[14,60]

Table 5   **Bronchoalveolar Lavage Data on Pneumoconioses—Pulmonology Research Centre**

Total Cells $\times 10^4$	Macrophages %	Lymphocytes %	Neutrophils %	Eosinophils %	CD4:CD8
20.2 ± 12.3	68.2 ± 22	22 ± 12.6	12.8 ± 15.2	0.2 ± 0.6	1 ± 0.9

BAL is a useful tool not only to clarify the immunopathogenesis of disease—a CD4 T cell profile with predominant TH1 differentiation eventually regulated by particular human leukocyte antigen (HLA)-DPB1 alleles[61]—but also in a diagnostic context, through the proliferative response of BAL lymphocytes to beryllium.

This lymphocyte transformation test, quantifying the proliferation of lymphocytes incubated with beryllium salts, is always positive in CBD patients if BAL lymphocytes are used, but positive in only 50% of cases if these cells are from the peripheral blood.[62] Thus BAL is the most sensitive tool to identify the disease. In a retrospective analysis of the last 20 consecutive patients with pneumoconioses with different etiologies submitted to BAL in the CPUC (results on Table 5), the authors found a less intense alveolitis compared with HP patients (Table 2) with a mixed lymphocytic/granulocytic alveolitis.

From the foregoing retrospective study a particular clinical case has been singled out that presents similar radiological findings to the patient mentioned in the case discussed earlier in which only ground-glass attenuation was shown, although it was less severe in expression and once again BAL was crucial for diagnosis.

A 64-year-old male patient, nonsmoker, who had worked in France for 9 years in contact with asbestos in a factory of thermal isolation materials, presented with complaints of progressive dyspnea and fatigue.

Lung function showed a restrictive ventilatory defect (FVC = 68.4%), with reduced diffusion capacity (TLCO = 65.2%).

HRCT scan revealed a ground-glass pattern, namely, in the inferior lobes (Fig. 6).

BAL showed the existence of alveolitis (cellularity of 34.8×10^4/mL) with 82.4% of macrophages, 16.3% lymphocytes, 0.8% eosinophils, and 0.5% neutrophils, and it was possible to identify ABs, in both the suspension (Fig. 7) and intercellularly, with the presence of a fiber phagocytosed by an activated macrophage (Fig. 8), as well as several ABs of 1.6/mL of BAL.

This clinical case illustrates a lung disorder related to occupation with clinical and functional repercussions and minor alveolar and radiological expression related to working with asbestos. To confirm

Figure 7 Asbestos fiber in bronchoalveolar lavage. MGG × 100.

Figure 6 High-resolution computed tomographic scan showing only minor ground-glass attenuation, in the left inferior lobe, on a subpleural localization.
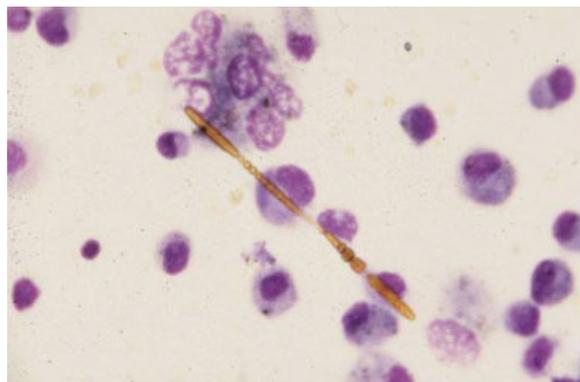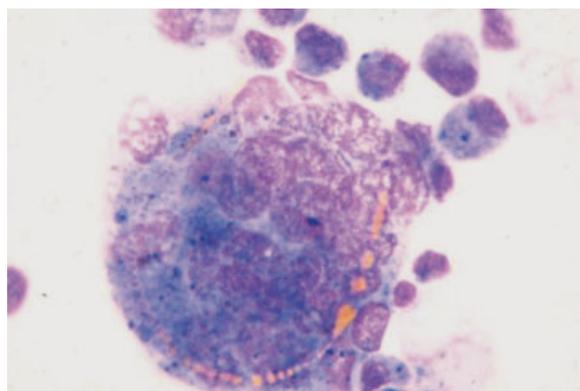

Figure 8 Bronchoalveolar lavage slide showing an asbestos fiber phagocytosed by macrophage. MGG × 100.

diagnosis, the identification and quantification of the ABs in BAL fluid were essential.

PERSPECTIVES

Therapeutic lavage is used in alveolar proteinosis and also, with smaller volumes, in some suppurative disorders.

Since Zhang et al[63] reported a delay in the development of disease in rats exposed to silica and submitted to this technique, whole lung lavage has a plausible role to remove inhaled particles in early stages of OLDs.

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