Chrysotile asbestos exposures can produce an alveolitis with limited fibrosing activity in a subset of high fibre retainer sheep

R. Bégin, A. Cantin, P. Sébastien

Chrysotile asbestos exposures can provide an alveolitis with limited fibrosing activity in a subset of high fibre retainer sheep. R. Bégin, A. Cantin, P. Sébastien.

ABSTRACT: Inhalation of fibrogenic mineral dust may enhance fibronectin (Fn) production by alveolar macrophages and increase fibroblast growth activity (FGA) in lung lavage fluid. To investigate the relationship of these changes to fibre retention and the development of asbestosis, we exposed 15 sheep to 100 mg Canadian chrysotile fibres in 100 ml saline, and ten sheep to 100 ml saline only, at ten day intervals. The animals were studied at 3 month intervals. At month 18, ten sheep had abnormal chest radiograph (category ≥1) (group B) and five had normal radiograph (category 0) (group A). Pulmonary function data indicated restrictive patterns of abnormalities in both groups, more severe in group B. Sheep in group A had bronchoalveolar lavage (BAL) cellularity and biochemistry comparable to controls; sheep in group B had significantly increased in total BAL cells (x2), macrophages (x2), neutrophils (x4) and eosinophils (x3), increased BAL lactate dehydrogenase and Fn, but FGA and procollagen 3 comparable to controls. Fibre retention was significantly increased in all exposed sheep and 2.5 x higher in group B vs group A despite similar exposures (70 intratracheal, 100 mg chrysotile infusions). Enhanced fibre retention in group B preceded the appearance of disease. This study confirms our earlier observation linking individual susceptibility to development of alveolitis to individual dust burden and provides evidence that the excess of fibre retention can be observed before detectable disease. In addition, we report a chrysotile-induced early alveolitis with incompletely expressed fibrosing activity at the time of initial radiographic detection. The intensity of the alveolitis is related to the degree of fibre retention. Eur Respir J. 1990, 3, 81-90.

Individual susceptibility of humans to development of disease is well recognized. In humans chronically exposed to asbestos dust inhalation at work, it is well documented that, for a given level of exposure, only a fraction of the workers develop asbestosis [1-6]. To investigate the so-called "individual susceptibility factor", immunological background, lung structure and lung clearance capacity have been considered.

Reports on human immunological histocompatibility in asbestos workers, including our own recent work [7, 8], have failed to identify definite markers for susceptible individuals. Anatomical structure characteristics of the major airways have been related to risk factors in adverse pulmonary response to asbestos exposure [2], and these characteristics may influence mucociliary clearance of the asbestos deposited in the lung.

Alveolar dust clearance in humans has not been directly studied as a determinant factor for individual susceptibility to asbestosis. However, lung tissue fibre burden has been found to be increased in asbestos workers compared to the general population [9]. Asbestos workers with isolated airway disease have twice the lung fibre content of workers without the airway disease [10] but only 50% of the fibre content of patients with asbestosis [11-14]. Analyses of bronchoalveolar lavage (BAL) fibre content of workers with asbestosis were also found to be significantly higher than values found in exposed workers without asbestosis [15, 16].

Since exposure levels cannot be precisely determined in human studies, it is not possible to compare groups of workers with similar exposure but different disease activity. Furthermore, the currently available techniques for measurement of fibre deposition and clearance using radioactive fibres evaluate primarily the rapid phase of dust clearance and its validity in reflecting the late slower phase of dust clearance, or overall clearance, have not been established.

Alternatively, fibre retention in the bronchoalveolar space, as determined by BAL, has been documented in humans [17, 18] and in our sheep model [19] to reflect
lung tissue retention. Thus, with this approach we estimated lung tissue fibre retention and related it to other phenomena in the sheep exposed to chrysotile dusts.

Our sheep model of asbestosis, which utilizes multiple intratracheal injections of the chrysotile dust as a means of exposure, has reproduced the heterogeneity of lung tissue response previously observed in humans [1-6] and other animal models [20, 21]. This model made it possible to establish that susceptible sheep were retaining more asbestos fibres [22, 23]. The levels of dust retention in our initial studies were measured only after disease was established.

The present experiment was therefore designed to test the above hypothesis of a dust retention related individual susceptibility. Thus, we obtained repeated measurements of BAL fibre content during the course of induction of experimental asbestosis.

An additional objective of this study was to characterize the fibrogenic activity of the early chrysotile-induced alveolitis in the susceptible sheep, at time of initial radiograph detection, and to relate the individual response of lung tissue to the degree of retention of the chrysotile fibres.

Methods

Animals. Twenty five sheep weighing 25-40 kg were used in this study. They were prepared and accustomed to the pulmonary techniques as previously reported [22, 23].

Experimental design. The flock was divided into a group of ten sheep exposed to phosphate buffered saline (PBS) only and a group of 15 sheep exposed to 100 mg UICC Canadian chrysotile asbestos fibres in 100 ml PBS every ten days. These fibres were relative uniform and well characterized, 92% being less than 0.25 μm in diameter and 20 μm in length. Exposures were carried out after nasotracheal intubation via repeated slow infusions of the suspension in the trachea at ten day intervals throughout the 24 month study.

Sequence of analyses. The animals were studied prior to exposure and at three month intervals thereafter by chest radiographs (CR), pulmonary function tests (PFT) and BAL analyses. Transthoracic lung biopsies were not obtained as we have previously correlated the histopathology of the early radiographic changes in this model [24-27].

Chest radiograph. Each sheep was positioned on a mobile cart with a wooden board and a grid cassette under the thorax. The X-ray source was placed at a 30° caudal angle, 2 feet from the cassette. The intubated animal was held at total lung capacity (TLC) using a giant syringe, and radiographs were taken at exposure factors 80 kV, 20 mAs, and 0.02 s. Each radiograph was scored according to the International Labor Organization (ILO) classification of radiographic profusion of parenchymal opacities [28]. This classification recognizes the existence of a continuum of change, from no opacity to the most advanced category. The scores were converted to a linear scale of 0-10 (12 categories) as follows: ILO grade 0/- (clearly normal) and grade 0/0 (normal after close examination) = 0 on the linear scale; 0/1 = 1; 1/0 = 2; 1/1 = 3; 1/2 = 4; 2/1 = 5; 2/2 = 6; 2/3 = 7; 3/2 = 8; 3/3 = 9; 3/4 = 10. In the ILO classification, four categories are defined on the basis of these same profusion scores: category 0 = profusion scores 0/-, 0/0, and 0/1; category 1 = profusion scores 1/0, 1/1, and 1/2; category 2 = profusion scores 2/1, 2/2, and 2/3; category 3 = profusion scores 3/2, 3/3, and 3/4. The radiographs in category 0 are generally considered normal whereas those in category 1 or above are definitely abnormal.

Pulmonary function tests. The methods used in PFT assessment of the sheep have been published previously [23-26]. Briefly, transpulmonary pressure was monitored with a naso-oesophageal 7 ml balloon catheter and an airway catheter connected to a Hewlett-Packard 270 differential pressure transducer (Hewlett-Packard, Waltham, MA). Gas flow at the airway opening was measured by connecting theuffed endotracheal tube to a Fleisch no. 2 pneumotachograph (Dyna-sciences, Blue Bell, PA) attached to a flow integrator recorder system and a Mink data processing system (Digital Equipment, Montreal, Quebec), for on-line analysis and storing of the data. Each PFT measurement was obtained after three inspiratory capacity measurements at a constant volume; TLC was defined as the lung volume at a transpulmonary pressure of +35±5 cmH₂O, and residual volume RV was defined as the lung volume at a transpulmonary pressure of -35±5 cmH₂O. The static expiratory lung compliance (Cslt) was determined by multiple-step syringe deflation between TLC and functional residual capacity (FRC). Diffusion capacity (DLco) was obtained by a passive rebreathing method using a gas mixture of 10% helium, 0.30% carbon monoxide, and 21% oxygen in nitrogen, a Collins catherometer (Warren E. Collins, Braintree, MA) and a Beckman infra-red carbon monoxide analyser (Beckman Instruments, Fullerton, CA). For the DLco test, the animals were passively ventilated with a 2.5 l syringe at a rate of 30 breaths-min⁻¹ with a 1 l tidal volume.

Bronchoalveolar lavage and cell analysis. The techniques in BAL procedures and analyses have been described previously [22, 23]. The BAL effluent was passed through four layers of cheesecloth to remove mucus, and the cells were pelleted by centrifugation. Cells were counted in a haemocytometer, and cell viability was determined by the trypan blue exclusion technique. Cytocentrifuge smears served to identify the cellular populations recovered with the Wright-Giemsa stain.

BAL proteins and enzymes. In the supernatant, albumin was determined by the immunochemical method of Killingsworth and Savory [29], with a laser nephelometer (Behring LK modular system, Hoechst Behring, Frankfurt, DDR) using specific antiseraum raised in...
rabbits (Cappel Lab. Inc., Downington, PA). The activity of lactate dehydrogenase (LDH), an indicator of cytoplasmic toxicity, was measured by spectrophotometric methods [30].

**BAL matrix constituents.** To assess interstitial lung matrix changes we looked at the fibronectin accumulations in BAL fluid. We also measured the production of fibronectin by BAL cells in culture [31, 32]. To determine cellular production of fibronectin, BAL cells were incubated without adherence step in 24 well culture plates (Linbro Chemical Co., New Haven, Conn.) in Dulbecco’s modified Eagles medium (DMEM, Gibco Diagnostic Laboratories, Grand Island, NY) containing 100 U·ml⁻¹ penicillin and 100 µg·ml⁻¹ streptomycin, at a density of 2x10⁶ cells·ml⁻¹, for 1 h at 37°C, 10% CO₂. At the end of the incubation period, the supernatants were centrifuged (500xg, 10 min) and stored at -20°C before fibroblast measurement. Fibronectin was determined by direct enzyme-linked immunosorbent assay (ELISA) technique adapted from Rennard et al. [31]. Briefly, 96 well polystyrene plates (Costar, Cambridge, Mass.) were coated with gelatin (500 µg per well) at 20°C for 12 h, rinsed three times with PBS, and samples of BAL supernatant were placed in each well at 20°C for 2 h. The plates were then washed and the concentration of fibronectin was determined by using antisheep fibronectin antibodies raised in rabbits and peroxidase conjugated goat antirabbit immunoglobulin G (IgG) (Boehringer Mannheim Biochemicals, Mannheim, DDR). Using the ELISA technique, the alveolar macrophages are the only BAL cells producing significant amounts of fibronectin [32].

**Fibroblast proliferation assay.** Since activated alveolar macrophages can release increased amounts of molecules that are mitogenic for fibroblasts [33, 34], we tested the release of fibroblast growth factors from lung inflammatory cells. Lung inflammatory cells were suspended in Dulbecco’s modified Eagles medium (DMEM) containing 100 U·ml⁻¹ penicillin and 100 µg·ml⁻¹ streptomycin at a density of 10⁶ cells·ml⁻¹ and incubated at 37°C, 10% CO₂ for 18 h. The adherence step was omitted here as macrophages were evaluated by analysis of variance for experiments and BAL cellularity of individual sheep with reference to the 95% confidence interval of control sheep also posed sheep and these changes were accentuated compared to saline exposed sheep. When a significant effect was detected, a Kruskal-Wallis test was used to determine which group means were significantly different (p<0.05) [36]. Analyses of correlation of fibres in BAL to other parameters of disease activity were performed with the Spearman correlation rank test [36].

**Results**

**Subsets of sheep.** At month 12, the group of 15 sheep exposed to chrysotile had significant changes in radiographic score, static lung compliance, vital capacity and arterial oxygen tension (Pao₂) compared to saline exposed sheep and these changes were accentuated thereafter. Analyses of individual results of the 15 asbestos exposed sheep showed that these changes were the effect of disease limited to ten of the 15 sheep. The chest radiograph was abnormal in ten. Analysis of lung functions and BAL cellularity of individual sheep with reference to the 95% confidence interval of control sheep also clearly separated the diseased subset.
We therefore divided the group of chrysotile exposed sheep into two subsets: a subset of five sheep with normal chest radiograph (0/0 or 0/1 score) (subset A) and a subset of ten sheep with abnormal chest radiograph (1/0 or above score) (subset B). In this model of asbestosis, histopathological changes associated with early changes in chest radiograph have been reported in detail [25-27] and briefly consist of a diffuse peribronchiolar macrophagic alveolitis extending into the adjacent interstitium and alveolar spaces.

**Bronchoalveolar exposure index.** This index is a time integral of fibre estimated to be retained in the bronchoalveolar space. It is expressed as number of fibres·μl⁻¹ per month. Results are presented in table 1. For the two groups of sheep, which received the same exposures, the bronchoalveolar exposure indices were not significantly different.

<table>
<thead>
<tr>
<th>Exposure</th>
<th>Control sheep</th>
<th>Sheep with normal radiograph</th>
<th>Sheep with abnormal radiograph</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bronchoalveolar exposure index (fibres·μl⁻¹ per month)</td>
<td>0</td>
<td>1601±590</td>
<td>1881±360</td>
</tr>
</tbody>
</table>

**Retention**

<table>
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<tr>
<th>Month</th>
<th>Fibre number fibres·μl⁻¹</th>
<th>Fibre length μ</th>
<th>Month</th>
<th>Fibre number fibres·μl⁻¹</th>
<th>Fibre length μ</th>
</tr>
</thead>
<tbody>
<tr>
<td>3</td>
<td>51.6±3</td>
<td>4.3±1.5</td>
<td>3</td>
<td>51.6±3</td>
<td>4.3±1.5</td>
</tr>
<tr>
<td>15</td>
<td>34.6±19</td>
<td>4.6±1.7</td>
<td>15</td>
<td>34.6±19</td>
<td>4.6±1.7</td>
</tr>
<tr>
<td>24</td>
<td>10.5±2.9</td>
<td>15.1±1.2</td>
<td>24</td>
<td>10.5±2.9</td>
<td>15.1±1.2</td>
</tr>
</tbody>
</table>

Bronchoalveolar exposure index is a time integral of fibre estimated to be retained in the bronchoalveolar space (fibres·μl⁻¹ per month). Results are geometric means±l.s. *: p<0.05 for sheep with abnormal radiograph vs sheep with normal radiograph. At month 24, mean fibre length was longer in exposed sheep but the difference between the two groups of exposed sheep was not significant.

**Lung functions.** In figure 1, we present the time course of changes in chest radiograph scores in each of the three groups of sheep in parallel with selected lung functions. It can be appreciated that, whereas lung functions of diseased sheep are clearly distinct from control sheep, the functions of chrysotile exposed sheep with normal radiograph remained between the normal and the exposed sheep. The fibre length also significantly changed in the two groups over time. In the chrysotile exposed sheep with normal chest radiograph, the mean fibre length·μl⁻¹ of BAL was 4.3±1.5, 4.6±1.7 and 15.1±1.2 at months 3, 15 and 24, respectively. In the chrysotile exposed sheep with abnormal chest radiograph, the mean fibre length was 3.2±1.4, 5.2±1.3 and 10.1±1.9 μM·μl⁻¹ at months 3, 15 and 24, respectively. In both groups, the average fibre length was significantly longer at month 24 only (p<0.05).

**Fibre retention.** BAL fibre analyses were obtained at months 3, 15 and 24 of the study (table 1). In the saline exposed sheep, fibre counts were always <0.2 fibres·μl⁻¹ and all were shorter than 1 μ in length. In the chrysotile exposed sheep with normal chest radiograph, fibre counts were 51.6±3, 34.6±19 and 10.5±2.9 fibres·μl⁻¹ at months 3, 15 and 24, respectively. In the chrysotile exposed sheep with abnormal chest radiograph, fibre counts were 84.1±2, 91.6±2 and 25.5±2.5 fibres·μl⁻¹ at months 3, 15 and 24, respectively. The ratio of fibre counts for normal/abnormal was 0.61, 0.37 and 0.41 at months 3, 15 and 24, respectively. Differences between the two groups were significant at p<0.05 at all times. A few asbestos bodies were observed in the radiographically abnormal groups. Lung resistance (Rt) at last point of study, month 24, was 0.24±0.08 kPa·l⁻¹·s⁻¹ in controls, 0.33±0.07 in subset A and 0.52±0.09 in subset B (p<0.05 for B vs controls). These changes in Rt were accompanied by significant reduction of effective lung compliance at 54 and 45% in controls and groups A and B, respectively. These values in both exposed groups suggested a peribronchiolar process as previously described in the sheep model [22-24].

**The alveolitis.** In figure 2, we present results of cellularity of lung lavage and in figure 3, the biochemistry and cell culture results. In the saline exposed sheep, there was no significant change over time in all parameters...
Fig. 1. - Results of chest radiograph scoring and lung function tests in the three groups of sheep. VC: vital capacity; $P_{aO_2}$: arterial oxygen tension; $DL_{CO}$: carbon monoxide diffusion capacity.

Fig. 2. - Time course of bronchoalveolar lavage cellularity in the three groups of sheep identified as in figure 1.
except for the increase in lymphocytes which has not been observed in our prior studies of the sheep model and remains otherwise totally unexplained. This was not observed in another control group of sheep evaluated during the same two year period (our silicosis study). The increase in lymphocytes in this study also caused total BAL cells to be slightly raised over time in controls.

In the chrysotile exposed sheep with normal chest radiograph, group A, values of cellularity were comparable to saline exposed sheep without significant changes in lymphocytes over time which remained at 10±2% of total BAL cells.

In the chrysotile exposed sheep with abnormal chest radiograph, group B, there were significant increases in total BAL cells, macrophages, eosinophils and neutrophils at month 12 and thereafter, compared to controls and group A. Furthermore, BAL albumin was slightly but significantly increased 50-70% above controls with parallel increases in IgG and IgM (data not shown) and more accentuated (300% of controls, month 18) increases in BAL lactate dehydrogenase. Fibronectin (Fn) production by macrophages in culture remained below detection level (<2 ng·10⁶ cells per 24 h) in controls throughout the study. In group A, Fn did not differ from controls. Fn in group B was increased above controls after month 12 and accumulated to values 300% of controls in the BAL supernatant of the sheep in group B. These changes occurred in the absence of significant increase in BAL procollagen 3/albumin ratio or fibroblast growth activity.

**Fibre retention correlates.** Analysis of correlation of fibre retention to other parameters of disease activity documented a significant relationship between individual fibre retention and radiographic disease (p<0.01), cytotoxicity (LDH in BAL) (p<0.01), fibronectin production by alveolar macrophages (p<0.01), increased cellularity of BAL (p<0.02) and loss of vital capacity (p<0.05). These associations strengthen the concept that, for a given chrysotile exposure, the intensity of disease process directly relates to the individual fibre retention.

**Discussion**

The present study reproduced the heterogeneity of lung tissue response previously observed in sheep chronically exposed to chrysotile [22, 23], in asbestos workers [1-6] and in other animal models of asbestosis [20, 21]. The sequential analyses of BAL fibre content, an index of alveolar dust retention [15, 17, 23, 37] documented the following observations which have not previously been reported. Firstly, given that exposure was continued throughout the 24 months of the study at the same exposure level (one injected dose per ten day interval), both groups of chrysotile exposed animals had a gradual decrease in alveolar dust retention which implies either
an enhancement of the alveolar clearance up the tra-cheobronchial tree or a larger transfer to the interstitial space. Secondly, fibre length significantly increased after month 15 in both groups of exposed sheep. The observation of a gradual decrease may be particular to the model and/or mode of exposure, but it also concurs with observations by Wagner et al. [20] on rat asbestososis, and Rowland et al. [38] and Sébastien et al. [39] in human asbestososis. The enrichment in long fibres has also been documented [39]. This enhanced alveolar clearance was observed in both groups of sheep with or without disease. Thirdly, the animals with asbestososis not only retained significantly more fibres but a significant excess of fibre accumulation could be detected before any abnormality of pulmonary radiograph, function or bronchoalveolar lavage. This observation therefore documented that this high retention state preceded the development of the disease. A link to individual susceptibility for disease development can, therefore, be firmly considered.

Our study is also of interest as it further characterized the early stage of asbestososis at the time of initial radiographic recognition of the disease process. Previous studies in humans [4, 40-42] and in several animal models, including sheep [22-27, 43-52] have clearly documented that lungs chronically exposed to asbestos or other mineral dusts have an excessive early accumulation of macrophages in the lower respiratory tract.

This initial macrophage alveolitis may not necessarily lead to interstitial lung fibrosis. Indeed, it has been documented in long-term asbestososis and silica workers [53] that the macrophage population of the lower respiratory tract may be expanded in the absence of interstitial lung fibrosis disease; in the animal model, it is a well-recognized early lung tissue reaction to inert dust particles which usually disappears without residual scar in the months after exposure cessation [54, 55]. This type of "transient macrophagic alveolitis" has been observed following latex bead, carbon, graphite, and particulate carborundum exposures [54-56]. Pathological characteristics of this lesion are its absence of distortion of the normal histological lung structure and the clearance of the phagocyte accumulation after cessation of exposure [54-56].

In terms of cell biology of the macrophage, we should ask what differentiates the macrophages of the transient dust alveolitis from that of the normal lung or that of a fibrosing alveolitis? Firstly, the macrophage of the normal lung is a permanent component of the lung defence mechanisms which, under normal healthy conditions, is in a quiescent state, producing minimal amounts of secretions [57, 58].

The deposition of the particles in the lower respiratory tract initiates in situ chemotactic activity for the macrophage which accumulates at the site of particle deposition [51]. Basal metabolic activity is increased, it displays pronounced ruffling of surfaces, phagocytosis and cell size increases, function is enhanced and, usually within hours, most particles deposited are phagocytosed [48-50]. If the particle or fibre phagocytosed is "inert", limited secretory activity of the alveolar macrophages occurs, the macrophage alveolitis is transient with little or no lung tissue scaring [55-57]. Under the circumstances of exposure to inert dusts, alveolar macrophages rapidly phagocytose the vast majority of particles but they do not increase secretion of molecules known to contribute to the pathogenesis of chronic lung disease [54-56].

Exposure of the lower respiratory tract to fibres or particles known to cause lung injury, bioactive, can lead to the following lung tissue responses:

1. The no-retention reaction: most fibres appear to be cleared away from the lung tissue rapidly without significant lung tissue scaring. This is the case in several experimental conditions of low exposures to chrysotile [59]. It is also the case in some long-term asbestososis workers which may be estimated in our clinical work to be up to 50% of all workers [4-6]. This has been observed in the animal model [59] and in long-term chrysotile miners [10].

2. The low-retention reaction: fibre retention is low because of the so-called "individual susceptibility factor", lung tissue reaction is limited to the site of deposition where macrophages accumulate and eventually are replaced by fibrotic scars limited to the distal airways [10, 23]. In this study it appears to be the case in our sheep in group A.

3. The high-retention reaction: fibre retention is significantly higher, lung tissue reaction is more intense, a diffuse macrophagic alveolitis can be recognized. The secretory activity of macrophages is enhanced. Oxidant production is increased [42], fibronectin production [22, 41, 42, 54], neutrophil chemotactic activity [60, 61] and fibroblast growth activity [42, 62] are increased, plasminogen activator [63] and interleukin 1 [64] are secreted at higher levels. If these secretions are sustained, diffuse lung damage occurs with the development of chronic interstitial lung disease. The latter observation has been particularly well documented in animal models.

This high-retention reaction is probably the case of individuals with interstitial lung disease associated with chronic inhalation of inorganic dust, asbestos in particular [41, 42]. The men with asbestosis have a high fibre content of their lung tissue [10-15] and a well-described fibrosing alveolitis where macrophages are clearly activated [5, 41, 42, 61]. This type of reaction has also been well characterized in animal models. In the sheep model [22, 27], this high retention reaction has all of the secretory characteristics reported in humans and becomes a progressive interstitial lung fibrosing disease even in the absence of further dust exposure. The animal models have demonstrated that these macrophage secretions needed to be sustained to initiate and maintain the development of a fibrosing lung process.

In this paper, we report on sheep which developed a relatively less intense alveolitis than in our previous experiments but, nonetheless, a diffuse reaction of the lung to chrysotile exposure. The sheep in group B had diffuse lung infiltrates; their BAL analyses documented an expanded cell population of the lower respiratory tract (fig. 2). Furthermore, there was evidence of enhanced secretory function of the cells; oxidant production was
increased, fibronectin production and accumulation was increased; LDH release was increased and albumin was accumulating slightly in the alveolar space. This alveolitis has been shown previously in the sheep model to enhance Gallium-67 lung uptake [4]. However, this reaction of modest intensity did not significantly alter fibroblast growth activity or increase procollagen 3 of lung lavage as previously documented in the presence of fibrosing lung disease [41, 42] and, thus, could at least "in theory", regress, leaving minimal lung damage.

This observation is of interest as we have previously documented enhanced prostaglandin E2, activity in the sheep model at the stage of early asbestosis [65], a prostaglandin which is capable of inhibiting fibroblast growth [66]. The prostaglandin E2 secretion by fibroblasts could be induced by alveolar macrophages and inhibit the fibrotic lung response [67].

This observation of an intermediate lung reaction between the "low- and high-retention reactions" occurring after exposures to dusts with known fibrogenic potentials is important as it describes a potentially reversible/non-scarring reaction if exposure is stopped. In addition, the present study documents that the chrysoile induced alveolitis at the time of initial radiographic detection has an incompletely expressed fibrosing activity. It could provide an explanation for the clinical observation of non-progression/regression of several cases of minimal asbestosis in humans.

References

ALVEOLITIS IN CHRYSOTILE ASBESTOS EXPOSURE


Les exposition à l’asbeste chrysotile peuvent provoquer une alvéolite avec activité fibrosante limitée dans un sous-groupe de moutons avec tendance élevée à rétention de fibres. R. Bégin, A. Cantin, P. Sébastien.

RÉSUMÉ: Des observation antérieures de notre laboratoire et d’autres institutions ont mis en évidence que l’inhalation de poussière mineure fibrogène peut stimuler la production de fibronectine par les macrophages alvéolaires (Fn) et augmenter l’activité de croissance des fibroblastes dans le liquide de lavage pulmonaire (FGA). Pour investiguer les relations entre ces modifications et la rétention de fibres, et le développement d’asbestose, nous avons exposé 15 moutons à 100 mg de fibres de chrysotile canadien dans 100 ml de solution saline à 10 jours d’intervalle, et 10 moutons à 100 ml de solution saline aux mêmes intervalles. Les animaux ont été étudiés à des intervalles de 3 mois par cliché thoracique, examen fonctionnel pulmonaire (PFT) et lavage pulmonaire (BAL). Au 18e mois, 10 moutons avaient des anomalies au cliché thoracique (catégorie 2) (groupe B), et 5 moutons avaient des radiographies normales (catégorie 0) (groupe A). Les épreuves fonctionnelles ont mis en évidence des types restrictifs d’anomalies dans les deux groupes, plus sévères toutefois dans le groupe B. Les analyses du BAL ont mis en évidence que les moutons du groupe A avaient une cellularity et un examen biochimique comparables à ceux des contrôle; par contre, les moutons du groupe B avaient des augmentations significatives du nombre total de cellules dans le BAL (x 2), des macrophages (x 2), des neutrophiles (x 4), et des éosinophiles (x 3). On notait en outre une augmentation de la lactate déhydrogénase du BAL, une augmentation de Fn, alors que FGA et le procollagène 3 étaient comparables aux contrôle. La rétention des fibres est significativement augmentée chez tous les animaux exposés, et 2,5 fois plus élevée dans le groupe B que dans le groupe A malgré des expositions similaires (70 infusions intra-trachéales de 100 mg de chrysotile). La stimulation de la rétention des fibres dans le groupe B a précédé l’apparition de la maladie. En conclusion, cette étude confirme nos observations antérieures, qui reliaient la susceptibilité individuelle à développer une alvéolite à la charge individuelle en poussière, et qui documentaient que l’excès de rétention de fibres peut être observé avant que la maladie ne soit détectable. En outre, nous avons mis en évidence une alvéolite précoce induite par le chrysotile avec une activité fibrosante incomplètement exprimée au moment de la détection radiographique initiale. L’intensité de l’alvéolite est en relation avec le degré de rétention de fibres.

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