Relationship Between Diffuse Pulmonary Fibrosis, Alveolar Proteinosis, and Granulocyte-Macrophage Colony Stimulating Factor Autoantibodies

Maurizio Luisetti MD, Pierdonato Bruno MD, Zamir Kadija MD, Takuji Suzuki PhD, Salvatore Raffa MD, Maria Rosaria Torrisi MD, Ilaria Campo PhD, Francesca Mariani MD, Ernesto Pozzi MD, Bruce C Trapnell MD, and Salvatore Mariotta MD

Extensive pulmonary fibrosis is a rare occurrence in pulmonary alveolar proteinosis. We report 2 cases that have interesting implications. A female patient was diagnosed with autoimmune pulmonary alveolar proteinosis that evolved over 7 years into diffuse fibrosis. In a male patient with diffuse fibrosis we incidentally detected electron microscopic features of alveolar surfactant accumulation and positive autoantibodies to granulocyte-macrophage colony stimulating factor. In the male patient we speculated that the pulmonary fibrosis might have been preceded by an asymptomatic phase of autoimmune pulmonary alveolar proteinosis, and that we should investigate the involvement of surfactant dysfunction in the pathogenesis of fibrotic lung disease. *Key words: pulmonary fibrosis; pulmonary alveolar proteinosis; alveolar surfactant; granulocyte-macrophage colony stimulating factor; surfactant catabolism; lamellar bodies; idiopathic interstitial pneumonia.* [Respir Care 2011;56(10):1608–1610. © 2011 Daedalus Enterprises]

Introduction

Pulmonary alveolar proteinosis is an autoimmune disorder caused by a dysfunction in surfactant catabolism due to the disruption of granulocyte-macrophage colony stimulating factor (GM-CSF) signaling in relationship to the presence of autoantibodies that neutralize GM-CSF.[1,2] A typical feature of pulmonary alveolar proteinosis is the integrity of alveolar septa, surrounded by an accumulation of amorphous, proteinaceous material within the air spaces.[3] Focal, mild fibrosis may be present, but extensive interstitial fibrotic disease is a rare occurrence in pulmonary alveolar proteinosis.[4] In this report we present data from 2 cases, with interesting implications.

Case Reports

Patient 1

Patient 1 was a female, aged 63 at diagnosis in 1999. She never smoked, but was occupationally heavily exposed to second-hand cigarette smoke, as a bartender. She complained of shortness of breath for 12 months, and when she was first examined, chest radiograph showed diffuse lung infiltrative disease with an alveolar filling pattern. Table 1 shows her lung-function data. High-resolution computed tomogram (CT) showed "crazy paving" pattern (Fig. 1A), and bronchoalveolar lavage (BAL) revealed features of accumulated amorphous, periodic-acid-Schiff-stain-positive material, so the diagnosis was idiopathic pulmonary alveolar proteinosis. During the observation period prior to whole-lung lavage, the patient had substantial im-

Drs Luisetti, Kadija, Campo, Mariani, and Pozzi are affiliated with Istituto di Ricovero e Cura a Carattere Scientifico Policlinico San Matteo Foundation, Pavia, Italy. Drs Bruno, Raffa, Torrisi, and Mariotta are affiliated with Unità Operativa Complessa Pneumologia and Diagnostica Cellulare, Medicina Clinica e Molecolare, Azienda Ospedaliera Sant' Andrea, Università di Roma La Sapienza, Roma, Italy. Drs Suzuki and Trapnell are affiliated with the Division of Pulmonary Biology, Department of Pulmonary, Critical Care, and Sleep Medicine, Cincinnati Children's Hospital Medical Center Cincinnati, Ohio.

The authors have disclosed no conflicts of interest.

Correspondence: Ilaria Campo PhD, Istituto di Ricovero e Cura a Carattere Scientifico Policlinico San Matteo Foundation, Via Taramelli 5, Pavia 27100, Italy. E-mail: i.campo@smatteo.pv.it.

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Table 1. Lung Function and Arterial Blood Gas Values

	Patient 1			Patient 2
	Pulmonary Alveolar Proteinosis Phase	Early Fibrotic Phase	Late Fibrotic Phase	
FEV_1, L/s (% predicted)	1.02 (71)	1.66 (114)	1.25 (77)	2.47 (71)
FVC, L (% predicted)	1.98 (109)	1.91 (111)	1.37 (70)	2.81 (63)
TLC, L (% predicted)	2.96 (77)	3.24 (88)	2.40 (63)	4.79 (61)
D_{LCO}, mL/min/mm Hg (% predicted)	699 (33)	627 (41)	547 (24)	697 (24)
P_{aO_2}, mm Hg	44	70	ND	45
P_{aCO_2}, mm Hg	33	41	ND	44

FVC = forced vital capacity
TLC = total lung capacity
D_{LCO} = diffusing capacity of the lung for carbon monoxide
ND = No data collected

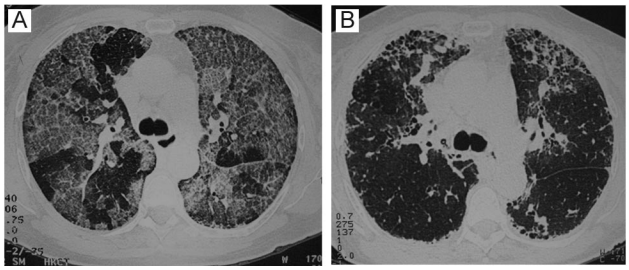

Fig. 1. High-resolution computed tomograms from patient 1. A: At diagnosis of pulmonary alveolar proteinosis (1999). B: At diagnosis of diffuse interstitial fibrosis (2006).

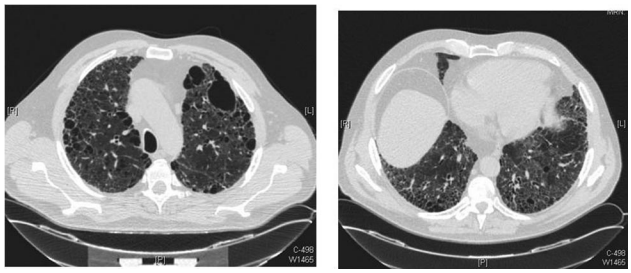

Fig. 2. High-resolution computed tomogram of patient 2, in February 2009, shows a reticular infiltrative process, architectural distortion, peripheral honeycombing, and traction bronchiectasis.

provement in lung function and gas exchange (P_{aO_2} on room air 77 mm Hg), so we postponed whole-lung lavage. Then the patient missed several scheduled visits.

A follow-up CT in March 2003 showed a decrease in ground-glass opacities, but alveolar septa thickening remained unchanged, and there was initial honeycombing in the upper lobes. The patient missed several other visits and did not have another radiograph for 30 months.

In January 2006 she returned and presented with increased dyspnea and cough, and a completely novel CT appearance (Fig. 1B). The crazy paving pattern was replaced by diffuse fibrotic changes, with subpleural honeycombing. Lactate dehydrogenase was within normal limits. Extractable nuclear antigens assay was negative, but antinuclear antibodies assay was positive, at a titer of 1:160. No other signs or symptoms of diffuse collagen disorders were detected. The GM-CSF autoantibodies concentration at this visit (the test had not been available previously) was extremely high (274 μg/mL, normal value < 3 μg/mL). Despite the resolution of the specific pulmonary alveolar proteinosis CT lung scan features, the BAL revealed foamy macrophages and a proteinaceous, amorphous, periodic-acid-Schiff-stain-positive sediment. She received oral prednisone and had initial improvement of symptoms and gas exchange, which remained stable after prednisone tapering. Subsequently, another CT revealed fibrotic changes and increased honeycombing.

In February 2008 she had a mild, transient pneumomediastinum, progressive lung-function impairment, and desaturation on exercise. In June 2008 she went on continuous oxygen. She underwent 3 additional admissions for acute respiratory failure, and the last of these, in November 2009, was complicated by stroke. She died 3 days after admission. The last serum GM-CSF autoantibodies measurement (June 2009) was 28 μg/mL.

Patient 2

A 61-year-old male was admitted with a history of progressive dyspnea over a 3-month period. He was a heavy smoker (25 pack/years), and for 30 years he worked in construction. Figure 2 shows the chest CT. Lung-function testing showed a restrictive pattern and respiratory failure (see Table 1). There were no features of collagen vascular disease. BAL recovered clear fluid with no amorphous material in the sediment. The total cell count was elevated (35.7×10^6), with 82% macrophages, 10% lymphocytes, 7% neutrophils, and 1% eosinophils, and the T cell CD4+ve/CD8+ve ratio was 0.8. Since an occupational exposure to inorganic dust or fibers was hypothesized as

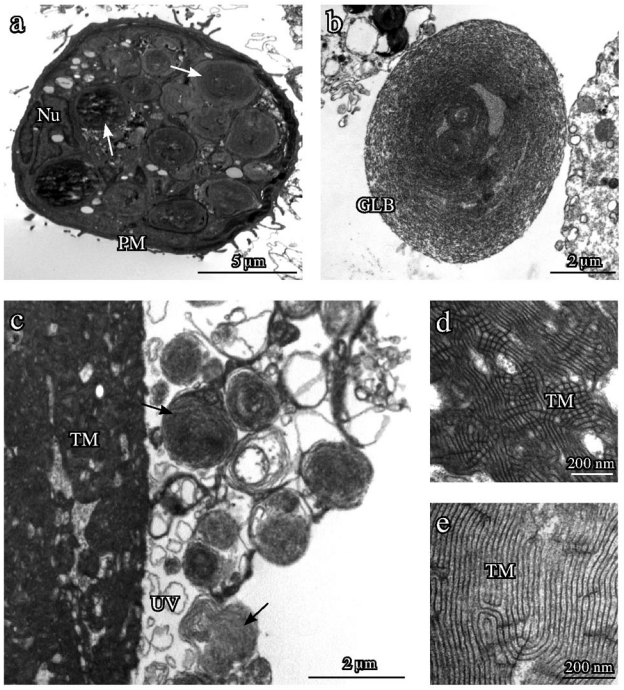

Fig. 3. Transmission electron micrographs of bronchoalveolar lavage fluid. A: Alveolar macrophage with intracellular accumulation of multi-lamellar bodies (arrows). Nu = nucleus. PM = plasma membrane. B: Giant multi-lamellar body (GLB). C: Large aggregate of intra-alveolar surfactant composed of tubular myelin (TM) figures, unilamellar vesicles (UV) and lamellar bodies (arrows). D and E: Details of the TM figures in the large aggregate.

the possible etiology of the fibrotic interstitial lung disease, we conducted electron microscopy of the BAL fluid (Fig. 3).

For conventional transmission electron microscopy we washed a pellet sample from the bronchoalveolar lavage 3 times in phosphate buffer and fixed with 2% glutaraldehyde in the same buffer (pH 7.4), for 2 hours, at 4°C, stained with uranyl acetate (5 mg/mL), dehydrated in acetone, and embedded in Epon 812 embedding resin. We examined ultrathin sections, unstained and stained with acetate and lead hydroxide, with an electron microscope (Morgagni 268D, FEI, Hillsboro, Oregon). The unexpected findings typical of pulmonary alveolar proteinosis prompted us to measure serum GM-CSF autoantibodies, which were present at 56 μg/mL (normal value < 3 μg/mL). The patient was discharged and given oral prednisone and oxygen supplementation. Sixteen months later he was stable.

Discussion

There are some clinical presentations where a relationship between surfactant and fibrogenesis has been established. For example, it is recognized that some inherited forms of surfactant homeostasis dysfunction, namely those related to ABCA3, SFTPA, and SFTPC gene variants, are associated with chronic interstitial lung disease.[5] Evolution of idiopathic (or autoimmune, as more recently defined) pulmonary alveolar proteinosis to fibrosis, such as that described as patient 1, is a rare but not exceptional occurrence.[6] However, patient 2 posed an intriguing situation: pulmonary fibrosis and GM-CSF autoantibodies, but absence of milky BAL fluid. This case points to the hypothesis that the pulmonary fibrosis was preceded by and linked to an asymptomatic phase of autoimmune pulmonary alveolar proteinosis, taking into account the high diagnostic specificity and sensitivity of the GM-CSF autoantibodies.

In fact, we have knowledge from a cohort, in Japan, of 233 patients with autoimmune pulmonary alveolar proteinosis, in whom 31% were asymptomatic and thus incidentally diagnosed.[7,8] Thus, we cannot exclude that some subjects diagnosed with diffuse fibrotic lung disease actually represented the end-stage evolution of a previous pulmonary alveolar proteinosis process. If we assume that the pulmonary-alveolar-proteinosis phase of patient 1 was asymptomatic, the CT performed during the fibrotic phase (see Fig. 1B) would not have indicated a surfactant catabolism dysfunction. An interesting, side finding in patient 2 was the relationship between the positive GM-CSF autoantibodies and the evidence of subclinical surfactant homeostasis dysfunction, as suggested by the electron microscopy data (see Fig. 3). Such a possible linear relationship merits further investigation. The findings from our 2 patients suggest involvement of a surfactant dysfunction in the pathogenesis of fibrotic lung disease.

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