

Autoimmune Pulmonary Alveolar Proteinosis Following Pulmonary Aspergillosis

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Abstract

A 59-year-old man, ex-smoker, was diagnosed with pulmonary aspergillosis according to chest radiography findings of a fungus ball and *Aspergillus fumigatus* detection in the bronchial lavage fluid. Two years after anti-fungal therapy, he was diagnosed with autoimmune pulmonary alveolar proteinosis (APAP) according to a crazy paving pattern in computed tomography scans of the chest, milky bronchoalveolar lavage effluent, and positive anti-granulocyte-macrophage colony-stimulating factor (GM-CSF) autoantibody in the serum. The patient ultimately died of respiratory failure caused by APAP four months after GM-CSF therapy commenced. *Aspergillus* infection may thus be associated with the onset and progression of APAP and tolerance to GM-CSF therapy.

Key words: pulmonary alveolar proteinosis, pulmonary aspergillosis, granulocyte-macrophage colony-stimulating factor

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Introduction

Autoimmune pulmonary alveolar proteinosis (APAP) is characterized by the accumulation of phospholipids and surfactant protein in the alveolar spaces (1) due to macrophage dysfunction caused by anti-granulocyte-macrophage colony-stimulating factor (GM-CSF) neutralizing antibody (2). Patients with APAP are immunocompromised because of functional disturbances of macrophages and neutrophils (3). Secondary infection (4, 5) with microorganisms, such as *Nocardia* (6), *Mycobacterium* (7), and *Aspergillus* (8, 9) species is also common in these patients. Infections have been reported to account for 18% of deaths in patients with APAP (4). We herein report a rare case of APAP in which *Aspergillus* infection may have caused the disease onset and/or progression of APAP.

Case Report

A 59-year-old man, ex-smoker, with a history of pulmonary tuberculosis complained of a dry cough for two months in November 2000. Chest radiography (Fig. 1A) and computed tomography (CT) (Fig. 2A, B) revealed a fungus ball in the pulmonary cavity of the left upper lobe that was caused by prior tuberculosis. According to these results, the patient was diagnosed with simple pulmonary aspergillosis (10). His serum was positive for anti-*Aspergillus* antibody. *Aspergillus fumigatus* was cultured from the bronchial lavage fluid collected from the left upper division of the bronchus. He was treated with itraconazole (100 mg/day) and his cough temporally improved; however, 2 years after the commencement of therapy (Fig. 1B), ground-glass opacity with a crazy paving pattern along with the gradual enlargement of the fungus ball was observed in chest CT scans (Fig. 2C, D) and the serum β -D glucan level increased to 101 pg/mL. Effluent obtained during the bronchoalveolar

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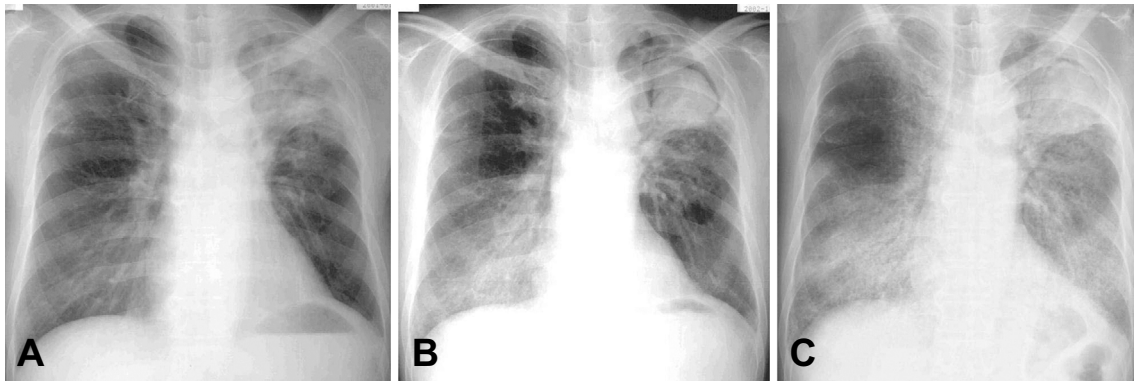


Figure 1. Changes in the chest radiographic findings. A fungus ball formed during previous tuberculosis appeared in the cavity of the left upper lobe (A). Two years after the diagnosis, the fields of the bilateral lower lung were found to exhibit ground-glass opacity (B), and complication of autoimmune pulmonary alveolar proteinosis (APAP) was diagnosed. APAP severity in the case increased 6 months after the diagnosis of APAP (C).

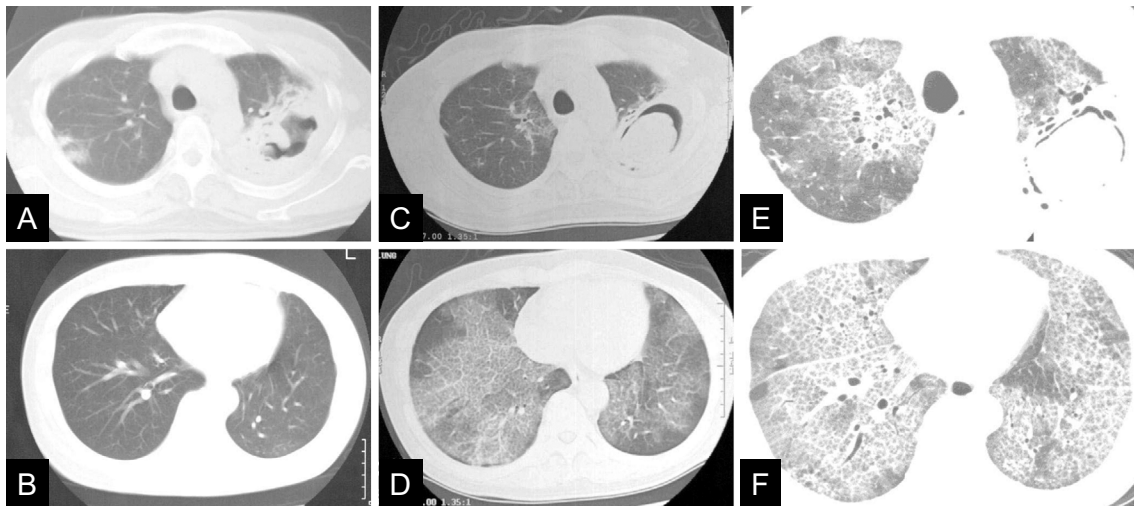


Figure 2. The chest CT scan. A thick-walled cavity was observed in the left upper lobe (A), and ground-glass opacity (GGO) was not observed at the diagnosis of *Aspergillus* infection (A, B). At the diagnosis of autoimmune pulmonary alveolar proteinosis (APAP), GGO suggestive of APAP appeared mainly in the lower lung field (D), and a fungus ball was present in the cavity (C). GGO with a crazy paving pattern extended to all lung fields, and the fungus ball enlarged in the cavity at commencement of therapy using granulocyte-macrophage colony-stimulating factor (E, F).

lavage appeared milky. Transbronchial lung biopsy specimens revealed alveolar wall thickening, alveolar type II epithelial cell hyperplasia, and periodic acid-Schiff stain (PAS)-positive microgranular materials in the alveolar spaces. The patient's serum was positive for anti-GM-CSF autoantibody (90.3 $\mu\text{g/mL}$, normal level: $<0.5 \mu\text{g/mL}$). The serum levels of Krebs von den Lungen-6 (KL-6) and surfactant protein-D (SP-D) were elevated to 5,700 U/mL and 583 ng/mL, respectively (normal levels: $<500 \text{ U/mL}$ and 110 ng/mL, respectively). The patient was subsequently diagnosed with APAP complicated by pulmonary aspergillosis.

Six months after the diagnosis of APAP (Fig. 1C, 2E, F), the patient's pulmonary function became severely disturbed, with a percentage predicted value of vital capacity and diffuse capacity of carbon monoxide at 39.5% and 20.4%, re-

spectively. An arterial blood gas analysis in the supine position under room air revealed a partial pressure of arterial oxygen (PaO_2) of 36.8 Torr. The disease severity of APAP was grade V, as defined by Inoue et al. (11). The serum KL-6 and SP-D levels were elevated to 24,500 U/mL and 1,180 ng/mL, respectively. Whole-lung lavage was deemed to be a contraindication due to active *Aspergillus* infection. The patient was treated by daily inhalation of 250 μg of GM-CSF for eight weeks; however, his hypoxemia did not improve. Although a daily subcutaneous injection of 250 μg of GM-CSF for three weeks was added to the treatment regimen, hypoxia and radiological findings did not improve. The intravenous administration of micafungin (150 mg/day) and inhalation of amphotericin B (5 mg/body) were initiated as antifungal therapy in place of itraconazole, which had been

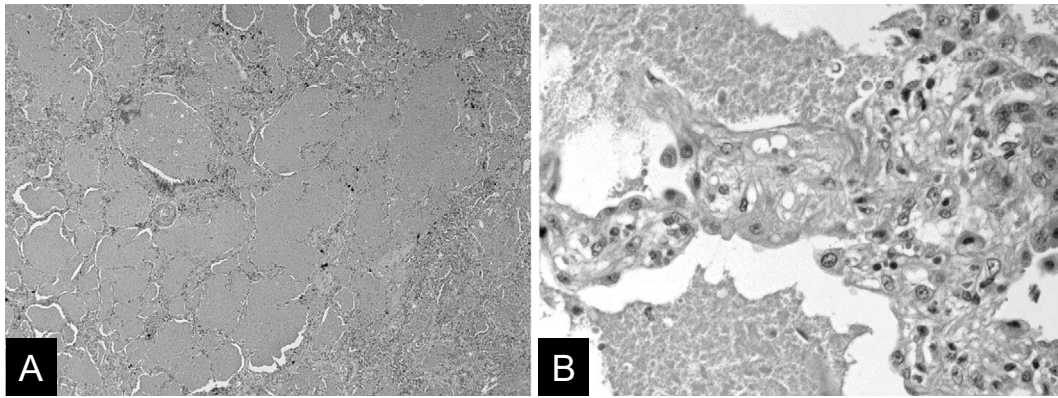


Figure 3. Pathological findings of necropsy specimens of the lung. The alveolar spaces were filled with PAS-positive microgranular material (A, B), and alveolar walls were lined with hyperplastic type II epithelial cells (B) (Hematoxylin and Eosin staining).

administered since the initial diagnosis of aspergillosis. The patient died of respiratory failure four months after the commencement of GM-CSF therapy.

Necropsy lung specimens revealed an intra-alveolar accumulation of PAS-positive material consistent with APAP, as well as neutrophil infiltration around bronchioles consistent with infectious bronchiolitis (Fig. 3); however, *A. fumigatus* was not detected in the small specimens. Additionally, interstitial pneumonia was not observed in the specimens. Hence, the chief cause of death was speculated to be a progression of APAP rather than *Aspergillus* infection.

Discussion

Immunodeficiency associated with APAP causes pulmonary aspergillosis (4, 5); however, in some cases, aspergillosis precedes APAP. Kobayashi et al. (9) reported that APAP in one patient following the onset of simple pulmonary aspergilloma resolved spontaneously after resection of the fungus ball. The resolution of PAP after the treatment of complicated rhinocerebral aspergillosis has also been reported (12). Hence, the pathophysiological association between *Aspergillus* infection and APAP is supported by these previous studies and aspergillosis may have affected the onset and progression of APAP in the present case.

The culture supernatant of aspergilli has been reported to suppress the macrophage function (13-15). Thus, aspergillosis could increase the deposition of microgranular materials in alveolar spaces, thereby worsening APAP by impairing phagocytosis by alveolar macrophages. The inhibition of macrophage function may be also associated with the disease onset of APAP. Anti-GM-CSF autoantibody is a pathogenic factor in APAP (16). However, low levels of anti-GM-CSF neutralizing antibody have also been reported to be present in the serum of healthy subjects and multiple myeloid functions may be regulated by GM-CSF concentrations (17). Therefore, in some situations, the elevation of serum levels of anti-GM-CSF neutralizing antibody to a critical threshold can lead to the development of APAP (17). *As-*

pergillus infection may lower the critical threshold for the concentration of anti-GM-CSF neutralizing antibody through its inhibitory effects on the macrophage function.

The production of anti-GM-CSF antibody may be stimulated in patients with *Aspergillus* infections. Th2 cells are activated in an animal model of aspergillosis (18, 19). This Th2 response is thought to be associated with autoimmune diseases by promoting the production of autoantibodies (20). Thus, *Aspergillus* infection may be an initiating factor for APAP in some cases.

The serum concentration of SP-D, which is reported to correlate with APAP severity (4, 21), has been found to increase in an *in vivo* murine model of aspergillosis (22) and in patients with allergic bronchopulmonary aspergillosis (23). The elevation of serum SP-D levels may reflect SP-D overproduction by alveolar epithelial cells, as aspergilli stimulate SP-D production by type II alveolar epithelial cells *in vitro* (18). Therefore, *Aspergillus* infection may also be a promoting factor for APAP by stimulating SP-D production by type II alveolar epithelial cells.

The tolerance to GM-CSF therapy observed in the present case may be due to variable effects of aspergilli. The resection of the fungus ball in the left upper lobe in this case may have reduced the severity of APAP, as previously described in the reports of Kobayashi et al. (9) and Lin et al. (12). It may also have improved the response to GM-CSF therapy. However, it was difficult to decide whether to perform resection of aspergilloma in the present case due to the progressive respiratory failure.

The hypothesis described above should be confirmed by clinical findings in other cases; however, it might be better that we suspect the complication of APAP if new ground-glass opacity (GGO) developed on chest CT in cases of *Aspergillus* infection. Whether anti-GM-CSF antibody is present in the serum of patients with *Aspergillus* infection and whether the change in anti-GM-CSF antibody level is associated with other clinical findings suggesting the onset of APAP should be clarified. These findings together may help to reveal the relationship between APAP and *Aspergillus* in-

fection. Furthermore, it may lead to improved methods for inducing remission of APAP cases that have become complicated due to *Aspergillus* infection and are resistant to GM-CSF treatment.

Author's disclosure of potential Conflicts of Interest (COI).

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