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【 CASE REPORT 】

Pathological and Radiological Correlation in Prolonged Myeloperoxidase Anti-neutrophil Cytoplasmic Antibody-related Diffuse Alveolar Hemosiderosis

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Abstract:
A 60-year-old woman with a 20-year history of myeloperoxidase anti-neutrophil cytoplasmic antibody (MPO-ANCA)-associated vasculitis visited our hospital due to productive cough and a low-grade fever for several weeks. Thoracic computed tomography demonstrated scattered tiny nodules, patchy consolidation, ground glass opacities, and thickening interlobular septa. On video-assisted thoracic surgery, those abnormalities were found to correspond to the accumulation of hemosiderin-laden alveolar macrophages (AMs) in the alveolar spaces and alveolar septa due to MPO-ANCA vasculitis. The radiological findings persisted for a further two years, indicating the possibility of persistent vasculitis in the lung or evidence of incomplete clearance of hemosiderin-laden AMs.

Key words: hemosiderosis, MPO-ANCA, thoracic computed tomography, hemosiderin laden alveolar macrophage

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Introduction

Hemosiderin-laden alveolar macrophages (AMs) in bronchoalveolar lavage fluid are usually considered a hallmark of alveolar hemorrhaging (1). Several quantitative diagnostic methods for the differential diagnosis of alveolar hemorrhaging have been described in previous reports (2, 3). However, the correlation between the radiological and pathological hemosiderosis findings has been studied very little.

We herein report a unique case in which a patient with massive pulmonary hemosiderosis due to myeloperoxidase anti-neutrophil cytoplasmic antibody (MPO-ANCA)-associated vasculitis was successfully evaluated by both thoracic computed tomography and pathological findings.

Case Report

A 60-year-old woman suspected of having miliary tuberculosis was referred to our hospital due to productive cough with a low-grade fever that had persisted for two weeks. She had been diagnosed with MPO-ANCA-associated vasculitis complicated with rapidly progressive glomerulonephritis 20 years prior to visiting our hospital. Since her diagnosis, the patient had been treated with maintenance hemodialysis and prednisolone (10 mg/day) without any symptoms but had experienced a persistently high MPO-ANCA serum titer ranging from 200 to 400 IU/mL for the past 2 decades. She had never smoked and had no history of illicit drug use or dust exposure.

On admission, her vital signs were normal except for tachycardia (118 beats/min), and a physical examination showed fine crackles posteriorly in the bilateral lower lung fields. Chest X-ray showed small nodules with faint infiltration that were noted mainly in the middle and lower lung fields (Fig. 1A). Thoracic computed tomography (CT) showed tiny nodules scattered throughout the lungs, especially in the bilateral lower lobes with grand glass opacities

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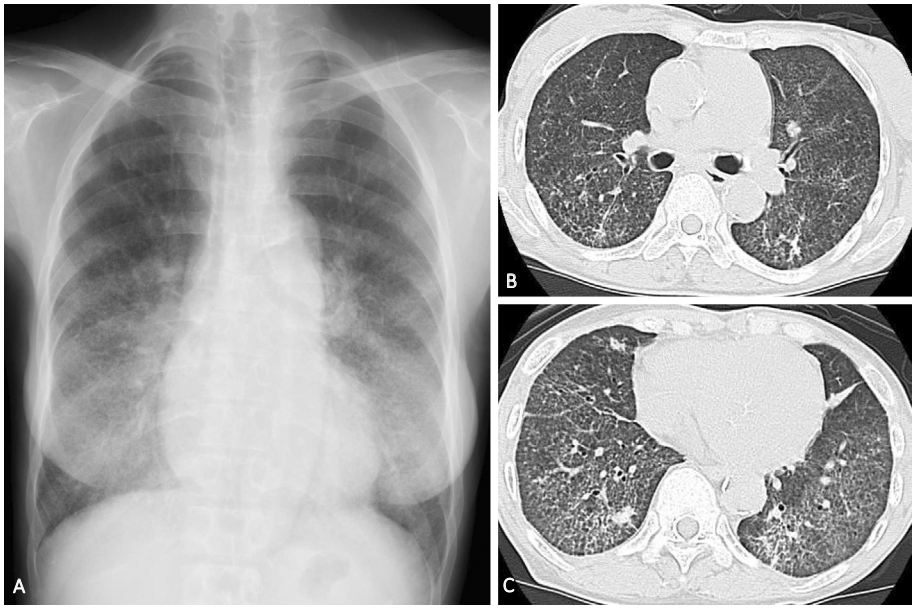

Figure 1. **Chest X-ray (A) on admission showed bilateral nodules with faint infiltration in the whole lung fields, especially in the middle to lower lung fields. Thoracic CT demonstrated scattered tiny nodules in the whole lungs (B, C), and patchy consolidation, ground glass opacities, and thickening of interlobular septa (C) were noted, particularly in the bilateral lower lung lobes.**

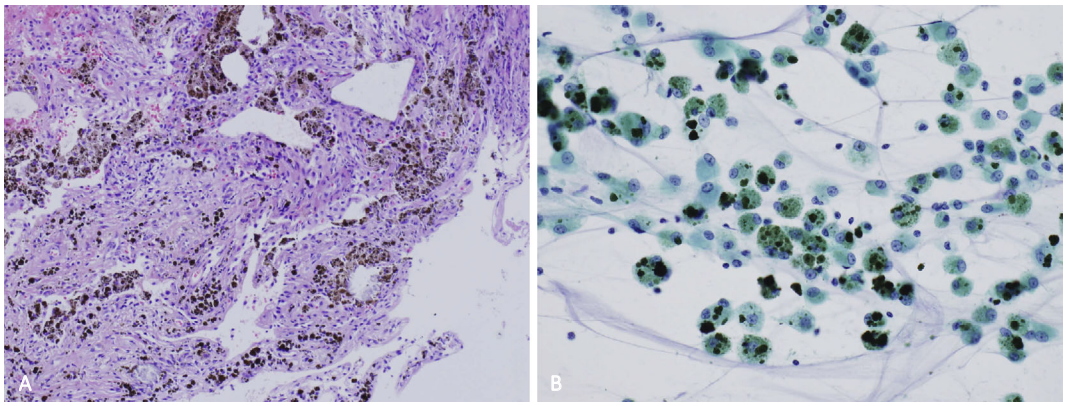

Figure 2. **Hematoxylin and Eosin staining of biopsied specimens obtained from B8 (A) revealed abundant hemosiderin-laden macrophages in the alveolar spaces. Papanicolaou staining of the bronchial washing fluid retrieved from B8 (B) showed numerous alveolar macrophages that contained the accumulation of intracytoplasmic hemosiderin, which was seen as faint, brown-colored, dense deposits.**

(Fig. 1B, C) and interlobular septal thickening (Fig. 1C). Several patchy consolidations were also noted in both lungs. A serum blood gas analysis revealed pH, 7.321; $PaCO_2$, 32.0 torr; PaO_2, 64.4 torr; HCO_3^-, 16 meq/L, and alveolar-arterial oxygen difference, 45.6 torr, findings that were suggestive of an impaired diffusion capacity. Pulmonary function tests demonstrated a restrictive pattern characterized by a vital capacity (VC) of 1.171 L, %VC of 52.8%, forced VC (FVC) of 1.13 L, forced expiratory volume % in 1 second (FEV 1.0%) of 0.861 L, and $\%FEV_1$ (G) of 76.1%. A serum laboratory examination revealed slight leukocytosis (white blood cell count: 9,500/μL) and high C-reactive protein (CRP; 18.7 mg/dL) and MPO-ANCA (148 IU/mL) values. The serum value of KL-6 (909 U/mL) but not SP-D (102 ng/mL) was high.

Urgent bronchoscopy was performed, and while no microbiological findings of *Mycobacterium tuberculosis* were found, the bronchoalveolar lavage fluid (BALF) obtained from B4b showed an elevated total cell count (0.9×10^5/mL), of which 3% were neutrophils, 40% were lymphocytes, 1% were eosinophils, and 56% were AMs. The CD4-to-CD8 lymphocyte ratio was 0.3. Hematoxylin and eosin (H&E) staining showed that all bronchial biopsied specimens obtained from B4, B8 (Fig. 2A), and B9 had abundant hemosiderin-laden macrophages in the alveolar spaces. Papanicolaou staining (Fig. 2B) of bronchial washing fluid re-

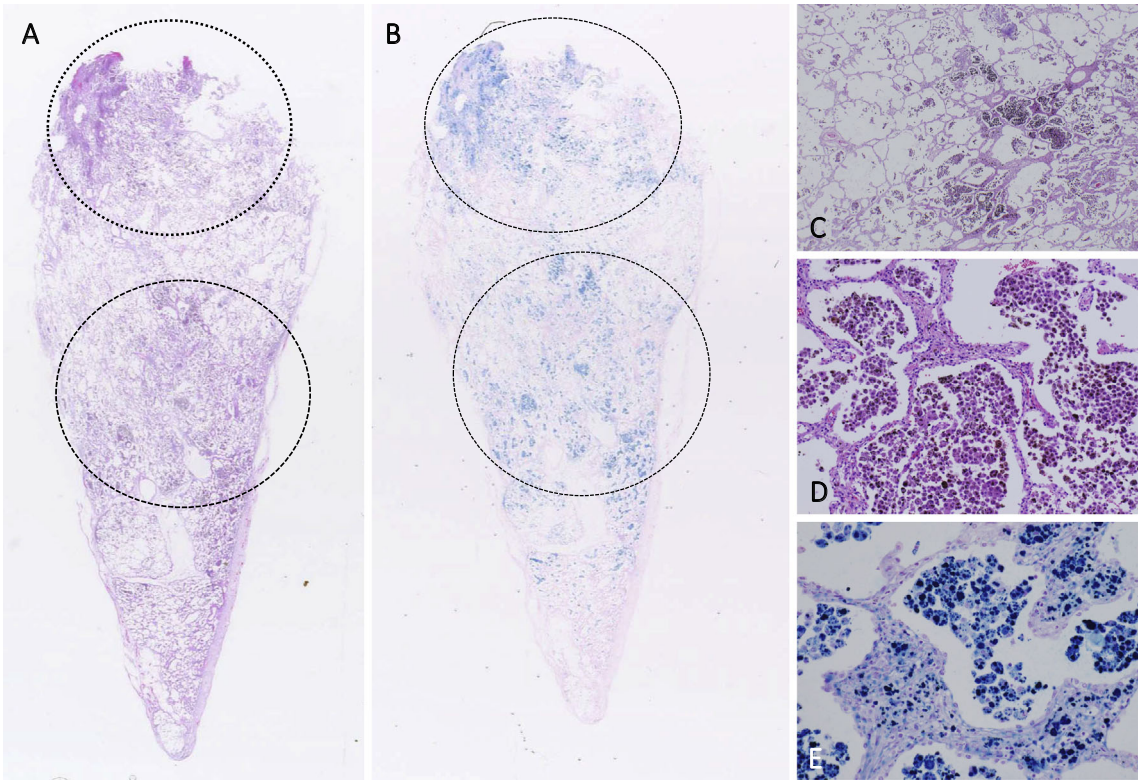

Figure 3. **A panoramic view of biopsied specimens from S9/S10 by video-assisted thoracic surgery revealed the presence of a mixture of dense (dotted circle) and faint deposits in the lung on Hematoxylin and Eosin (H&E) staining (A) and iron staining (B). H&E staining showed the accumulation of hemosiderin-laden AMs in the alveolar spaces (C, 100× magnification) that contained brown-col ored pigments (D, 400× magnification) and interlobular septal thickening (D). Iron staining showed AMs with blue granules in the cytoplasm (E). AMs: alveolar macrophages**

trieved from B8 showed numerous AMs that contained the accumulation of intracytoplasmic hemosiderin, seen as faint, brown-colored, dense deposits.

To confirm the diagnosis and explore the underlying illness, video-assisted thoracic surgery (VATS) was performed. Resected specimens obtained from the right border area of S 9/S10 (Fig. 3A) in the panoramic view of H-E staining demonstrated a complex of dense and faint components in the lung that mainly consisted of iron-containing substances (Fig. 3B). H & E staining showed patchy infiltration of hemosiderin-laden AMs in the alveolar spaces together with interlobular septal thickening in areas where the AMs were abundant (Fig. 3C, 100× magnification). These findings were confirmed by the pathological findings of both H & E (Fig. 3D, 400× magnification) and iron staining (Fig. 3E, 400× magnification), in which abundant AMs, engulfed in brown (Fig. 3D) or blue pigment (Fig. 3E), were apparent in the alveolar spaces and alveolar septa. Furthermore, a panoramic view of resected lung specimens stained with Elastica Masson Goldner (EMG) also showed a mixed component of dense and faint areas (Figure not shown). The intensity of interlobular septal thickening differed between the dense area and the faint area and seemed to depend on the severity of the accumulation of hemosiderin-laden AMs. Importantly, on H & E staining, a tiny nodule measuring 3 mm in size was noted (Fig. 4A). The nodule contained a small, scarred hematoma (Fig. 4B, 400× magnification) surrounded by fibrous tissue with destruction of the elastic lamina (Fig. 4C, 400× magnification) and visceral pleural thickening (Fig. 4A), which was indicative of an immunological disorder, such as collagen vascular disease (including vasculitis).

The patient developed scleritis immediately after the VATS biopsy. Based on these findings, the patient was diagnosed with MPO-ANCA-associated diffuse alveolar hemosiderosis. After the initiation of high-dose steroid treatment, her symptoms subsided, and her CRP and MPO-ANCA serum values normalized. These values remained well controlled over the following two years. However, chest X-ray taken two years later (Fig. 5A) still showed faint infiltration with small nodules in the middle to lower lung fields. In addition, thoracic CT still showed scattered tiny nodules (Fig. 5B, C) and ground glass opacity (GGO) with interlobular septal thickening (Fig. 5C) in both lower lungs, as previously recognized (Fig. 1).

Discussion

The pathological findings of hemosiderosis in idiopathic cases (4) and other diseases, including MPO-ANCA (5), have been studied very little. In this regard, the present case

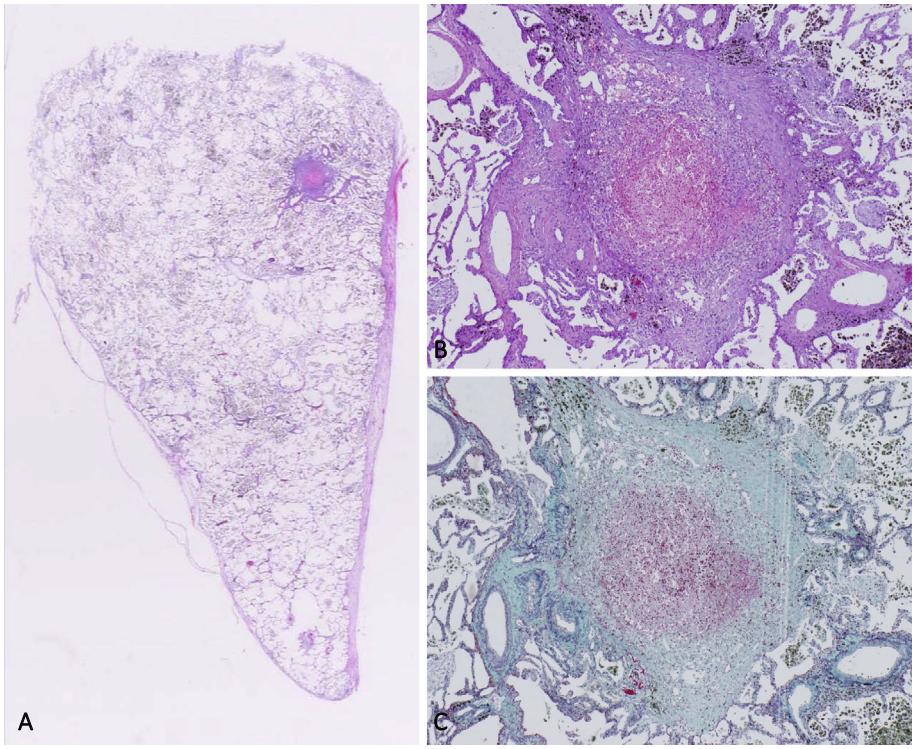

Figure 4. **Hematoxylin and Eosin staining in a panoramic view showed visceral pleural thickening with a tiny nodule measuring 3 mm in size (A) that contained a small, scarred hematoma (B, 400× magnification). Elastica-Masson staining showed fibrous tissue surrounding the nodule with destruction of the elastic lamina (C, 400× magnification), suggestive of vasculitis.**

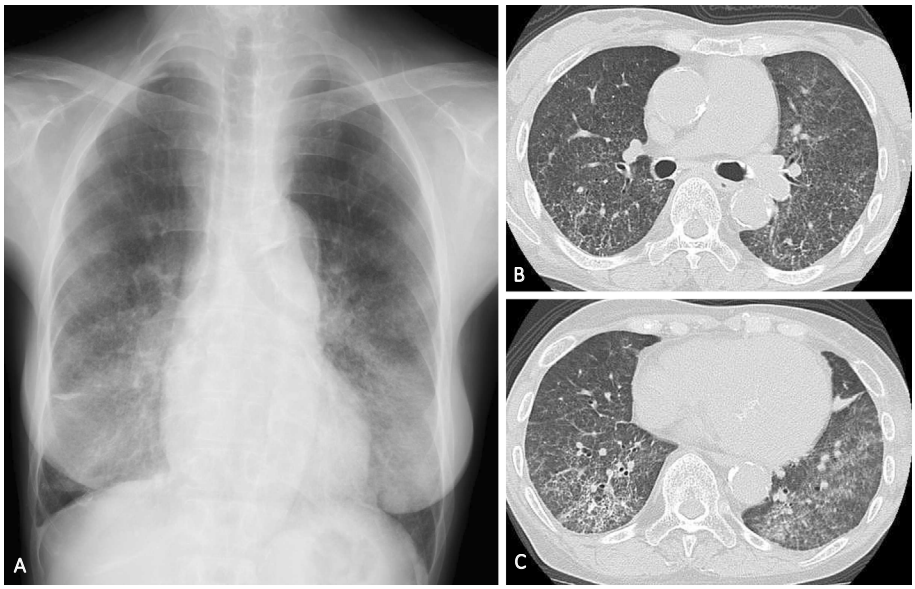

Figure 5. **Two years after the patient's first referral, chest X-ray (A) still showed small nodules and faint infiltration in the bilateral middle to lower lung fields. Thoracic CT depicted scattered tiny nodules in the whole lungs (B) together with ground glass opacities and thickening of the interlobular septa in both lower lobes (C).**

successfully confirmed that tiny nodules, patchy consolidation, and GGO corresponded to the accumulation of hemosiderin-laden AMs in the alveolar spaces. Furthermore, the infiltration of hemosiderin-laden AMs in the alveolar septa and alveolar spaces was attributed to the formation of GGO or interlobular septal thickening. To our knowledge, this is the first case with evidence showing a definite radiological-pathological correlation in hemosiderosis.

The onset of hemosiderosis could not be determined in this case, but small nodules in the lung had been identified

10 years prior to the patient's first referral to our hospital. In addition, an analysis of the BALF demonstrated that the cell differentiation was lymphocyte-predominant, suggesting a chronic status of the disease. Furthermore, two years after the diagnosis of hemosiderosis, the lung lesions appeared to be the same despite resolution of the patient's symptoms. This might be indicative of recurrent vasculitis in the lung or of hemosiderin-laden AMs within a single area over an extended duration without exacerbation of the vasculitis; in a clinical or radiological context, the present case was more likely a result of the latter hypothesis. Indeed, we previously reported a case with persistent scattered pulmonary nodules that corresponded to pulmonary calcification in the alveolar walls and interstitial space (6).

Little is known about the time course of hemosiderin formation and clearance from the human lung. Hemoglobin and intact erythrocytes in AMs are evidence of recent hemorrhaging (7), and previous reports have determined that AMs retrieved from healthy, non-smoking young adults can convert to hemosiderin-laden AMs within 72 hours following exposure to antibody-coated sheep red blood cells *in vitro* (2). Sherman et al. determined that total clearance of hemosiderin in humans to be possible within two to four weeks following acute hemorrhaging (8). However, it takes a long period of time (up to two months) for hemosiderin to be cleared in animal models (8), possibly because the clearance mechanisms for AMs become overwhelmed. It may therefore be difficult to differentiate between acute bleeding in the lung and chronic recurrent bleeding, as in the present case. However, the present case showed a correlation between the pathological and radiological findings with a focus on hemosiderin-laden AMs, and we discussed the process through which these unique radiological patterns might have been obtained.

Conclusion

Tiny nodules, patchy consolidation, and GGO on thoracic CT likely correspond to the accumulation of hemosiderin-laden AMs in alveolar spaces. The infiltration of hemosiderin-laden AMs in the alveolar septa and/or spaces can be attributed to the formation of GGO or interlobular septal thickening. This case study of hemosiderosis-associated MPO-ANCA is the first to report a definite radiological-pathological correlation.

The authors state that they have no Conflict of Interest (COI).

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