Pulmonary Immune Complex Deposition in Wegener's Granulomatosis*

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Two male patients with pulmonary manifestations of Wegener's granulomatosis are presented. One had an elevated rheumatoid factor, and both had elevated levels of immunoglobulin E. Both demonstrated characteristic necrotizing granulomatous lesions on light microscopy of lung tissue. Immunohistologic analysis of lung tissue demonstrated a granular deposition of immunoglobulin G and complement. Raji cell assay of sera demonstrated elevated levels of circulating immune complexes in the sera of the one patient tested prior to any therapy. These findings support the hypothesis that immune complex deposition contributes to the pathogenesis of Wegener's granulomatosis.

Wegener's granulomatosis is characterized by necrotizing granulomata of the respiratory tract, focal (often necrotizing) glomerulonephritis, and disseminated vasculitis involving both small arteries and veins. While these histologic changes are well recognized, their etiology remains obscure. Early reports emphasized a "hypersensitivity phenomenon" as a probable cause of the granulomatous and vascular changes, and recently, immune complexes have been implicated in their pathogenesis. Evidence for this includes the following: deposition of immunoglobulin in renal tissue, subepithelial electron dense deposits in kidney sections, and the presence of circulating immune complexes in the sera of these patients. In contrast, other authors failed to find evidence for immune complex deposition in renal tissue, making the role of immune complexes in the pathogenesis of Wegener's granulomatosis uncertain. To date, immunopathologic examination of lung tissue from Wegener's granulomatosis patients has not been reported.

Here, we report two patients with Wegener's granulomatosis and pulmonary involvement. Immunohistologic sections of lung tissue demonstrated the deposition of immunoglobulin and complement, and circulating immune complexes were present in the sera of the one patient tested prior to receiving therapy.

Case Reports

Case 1

A 25-year-old man was referred to the University of Colorado Medical Center in January, 1978 for a decrease in visual acuity. Limbal opacities, as well as scleral inflammation and anterior chamber cells were present. Prednisone, 80 mg/day, was begun.

After a six-month remission, he was re-admitted in late September with progression of the ocular findings and a three-week history of a productive cough. He was a chronically ill appearing young man who avoided the light. The conjunctivae and sclerae were inflamed with nodules in the sclerae at the limbal borders. The tympanic membranes were scarred and perforated and there was a mucous ulcer on the left side of the nasal septum. Basic immunologic parameters are listed in Table 1. The electrocardiogram showed right bundle branch block pattern. The chest roentgenogram demonstrated a large right upper lobe cavity and several other cavitating and non-cavitating lung nodules.

A biopsy of the nasal ulcer showed acute and chronic inflammation. He developed complete heart block and an open lung biopsy revealed a granulomatous, necrotizing vasculitis consistent with Wegener's granulomatosis (Fig. 1). Cyclophosphamide 200 mg/day, was added to the pred-

Table 1—Immunologic Parameters in Two Patients with Wegener's Granulomatosis

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<thead>
<tr>
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<th>Case 1</th>
<th>Case 2</th>
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<tbody>
<tr>
<td>Cryoglobulins</td>
<td>Negative</td>
<td>Negative</td>
</tr>
<tr>
<td>Hepatitis B surface antigen</td>
<td>Negative</td>
<td>Negative</td>
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<tr>
<td>Antinuclear antibody</td>
<td>Negative</td>
<td>Negative</td>
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<tr>
<td>Rheumatoid factor</td>
<td>1:1280</td>
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<tr>
<td>Total hemolytic complement</td>
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<td>Normal</td>
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<td>Serum protein electrophoresis</td>
<td>Increased 2 globulin</td>
<td>Increased 2 globulin</td>
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<tr>
<td>Immunelectrophoresis</td>
<td>Increased IgE</td>
<td>Increased IgE</td>
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CASE 2

A 65-year-old man complained of fevers, sweats, hemoptysis, and transitory chest pain for two weeks. A chest radiograph showed multiple bilateral lung nodules. He admitted to having sinusitis over the previous two years, but no other prior respiratory symptoms. Basic immunologic parameters are listed in Table 1. The chest roentgenogram demonstrated multiple bilateral lung nodules and less discrete lung infiltrates. Fiberoptic bronchoscopy gave no definite diagnosis and an open lung biopsy was done. Light microscopy showed necrotizing granulomata with foreign body type giant cells and an intense vasculitis in the vicinity of the granulomata.

The diagnosis of Wegener's granulomatosis was made and he began on 60 mg of prednisone and 150 mg of cyclophosphamide. Five months later, he was clinically well, and findings on the chest film were normal.

SPECIAL STUDIES

Immunohistologic Analysis of Lung Tissue

A portion of the tissue obtained at open lung biopsy was prepared for immunohistologic analysis. The methods are those previously described. Briefly, tissue was immediately frozen in Ames embedding medium and stored at −70°C. Four micrometer sections were cut, and the sections were exposed to fluorescein isothiocyanate conjugated goat antisera to human IgM, IgG, IgA, IgE, the third component of complement (C3), fibrinogen, and albumin. The antisera had previously been absorbed with mouse liver powder. The sections were examined on a Zeiss microscope with an ultradark-field condenser and halogen light source.

The specificity of a positive staining reaction was established by: (1) the absence of alveolar wall staining of normal control lung; (2) the presence of positive staining for IgG, IgM, and C3 in known positive lupus erythematosis control kidney sections; and (3) the absence of staining in positive sections after blocking binding sites with nonconjugated antisera.

Assay of Serum for Immune Complexes

Blood was obtained by venipuncture at the time of the open lung biopsy, and levels of circulating immune complexes were measured using the Raji cell technique. Briefly, the Raji cell is an immunoblastoid cell with surface receptors which bind antigen-antibody complexes containing complement. Raji cells exposed to test serum are incubated with radiolabeled antibody to human immunoglobulin. The radioactivity of cells exposed to test sera is compared to that of cells exposed to known amounts of heat-aggregated human gamma globulin, with human serum as a source of complement. Results are expressed as micrograms of aggregated human globulin equivalents per milliliter of serum (μg eQ AHG/ml).

RESULTS

Immunohistology

Immunohistologic sections from both cases are shown in Figures 2 and 3. Both demonstrated positive immunofluorescence for IgG and C3 in a granular pattern within alveolar structures, as well as medium-sized blood vessel walls. In contrast to the granular staining for IgG and C3, staining for fibrinogen was present in a linear pattern. Staining for albumin was negative indicating the lack of nonspecific transduction. Staining for IgA and IgM was also negative.
Circulating Immune Complexes

Case 1, whose serum sample was obtained after nine months of high dose corticosteroid therapy, had a normal level (<12 μg eQ AHG/ml) of circulating immune complexes. Case 2 had a level of 216 μg eQ AHG/ml.

COMMENTS

Increased rheumatoid factor activity has frequently been noted in patients with Wegener’s granulomatosis, and as in our patient, it parallels the activity of the disease. Interestingly, the rheumatoid factor activity in our patient (1:1,210) did not correspond to the levels of circulating immune complexes (<12 μg eQ AHG/ml), thereby disputing the suggestion of some authors that immune complexes in Wegener’s granulomatosis contain rheumatoid factor.

The elevated immunoglobulin E (IgE) levels, seen in both cases, were previously noted by Conn et al in five of seven patients. He suggested, with others, that IgE may be interacting with basophils and mast cells causing the release of vasoactive substances which allow immune complexes to deposit extravascularly.

Previous immunohistologic studies in patients with Wegener’s granulomatosis have been restricted to renal tissue. Several reports demonstrated immunoglobulin and complement in renal biopsy sections, while two others found no evidence for immune complex deposition. The positive immunofluorescence seen in our patients is the first report of immunohistologic analysis of lung tissue in Wegener’s granulomatosis.

The simultaneous presence of circulating immune complexes in one of our patients is also consistent with this hypothesis. Circulating immune complexes were present in Wegener’s patients in two previous reports. In addition, they were noted to fall rapidly with therapy, and this may explain the normal levels found in Case 1 after nine months of high dose corticosteroid therapy.

The deposition of immunoglobulin and complement in the lungs of two patients with Wegener’s granulomatosis and the simultaneous presence of circulating immune complexes in the sera of the one untreated patient implies that the deposition and/or presence of immune complexes contribute to the pathogenesis of this disease. Immune complexes deposited in the lung could mediate injury in several ways. The complexes, interacting with mononuclear cells and platelets, are capable of increasing vascular permeability and producing local edema. The complexes are also capable of activating complement with subsequent chemotaxis and polymorphonuclear leukocytes. The leukocytes are able to phagocytose some of the complexes, but in so doing release proteolytic enzymes that can injure surrounding tissue. In addition, the chemotactic factors themselves, especially C5a, can induce this enzyme release. Whether the antigen in the complexes is the inciting agent of the disease or an altered host protein, the deposition of the complexes in lung parenchyma can initiate a sequence of events leading to tissue destruction. The demonstration of this deposition supports the hypothesis that immune complexes participate in the pathogenesis of Wegener’s granulomatosis.

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