Serum Concentration of Soluble Interleukin-2 Receptor as a Sensitive Parameter of Disease Activity in Sarcoidosis*

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We investigated the clinical value of measuring serum concentrations of soluble IL-2R in monitoring sarcoidosis. Serum concentrations of soluble IL-2R were measured in 70 patients with sarcoidosis. The mean value for active untreated sarcoidosis was 1,143 ± 500 U/ml, while the normal range in 97 healthy control subjects was 80 to 300 U/ml. The mean value for active untreated sarcoidosis was significantly higher than that for dormant disease (333 ± 183 U/ml) or that for corticosteroid-treated patients (380 ± 151 U/ml). Serial changes in serum soluble IL-2R level were studied in cases of spontaneous remission or in corticosteroid-treated patients; a good correlation was noted between the changes in serum level of soluble IL-2R and clinical status. A positive correlation was noted between serum concentration of soluble IL-2R and serum ACE activity. These data confirmed that measurement of serum concentration of soluble IL-2R could be used in monitoring the disease activity in sarcoidosis. (Chest 1990; 98:1125-29)

Although sarcoidosis is self-limited in most cases, some instances of the disease are progressive and occasionally fatal.1,2 Therefore, monitoring disease activity is of great importance in the management of this disease.3 Disease activity of sarcoidosis currently has been assessed by clinical status, chest x-ray film findings, pulmonary function tests, gallium 67 scans,5 BAL findings,6 or serum ACE levels.6,12 Chest roentgenogram and BAL findings may reflect the grade of pulmonary involvement of this disease, but they may not reflect the total amount of granulomatous changes of sarcoidosis. Furthermore, difficulty in repetitive performance of BAL or gallium 67 scanning militates against its use in monitoring the disease activity of sarcoidosis. Serum activity of ACE, which can be obtained easily and is considered to reflect the total amount of sarcoid granulomata,9 has been used as a good marker of the disease, but the overlap of the enzyme level between some patients with sarcoidosis and healthy controls limits its value in regard to its sensitivity.

Immunopathologically, sarcoidosis is a chronic granulomatous disease characterized by local accumulation of activated T-helper/inducer cells.13-15 In this context, it is reasonable that parameters indicating T-cell activation may reflect disease activity of sarcoidosis. Recently, it has been reported that the serum concentration of soluble IL-2R is elevated in patients with diseases known to be associated with T-cell activation, such as T-cell leukemia,16 autoimmune disease,17 tuberculosis18 and leprosy.19 The serum level of soluble IL-2R was also shown to be elevated in sarcoidosis,20 presumably reflecting the activated T-lymphocytes in this disease.21 We measured serum concentrations of soluble IL-2R to estimate its clinical usefulness in monitoring sarcoidosis.

Materials and Methods

Patient Population

Seventy patients with sarcoidosis were enrolled in this study (Table 1); 29 were males and 41 females with a mean age of 43.9 ± 15.1 years. At the time of diagnosis, all of the patients had typical clinical features of sarcoidosis and the diagnosis was made.

Table 1—Study Populations

<table>
<thead>
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<th>Type of Sarcoidosis</th>
<th>Active</th>
<th>Dormant</th>
<th>Treated</th>
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<td>8</td>
</tr>
<tr>
<td>Age (yr)</td>
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<td>42 ± 14</td>
<td>47 ± 17</td>
</tr>
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Assay for Soluble IL-2R in Serum

Measurement of serum concentrations of soluble IL-2R was performed by using commercially available Celfree Interleukin-2 Receptor Bead Kit (T Cell Sciences Inc, Cambridge, MA). This assay system is a two-site, solid-phase enzyme immunometric assay which employs two noncompeting monoclonal antibodies to the human IL-2Ra chain. Briefly, each 50 μL of serum sample was incubated with a plastic bead coated with a monoclonal antibody directed against one epitope on the IL-2R molecule, and with a horseradish peroxidase-conjugated monoclonal antibody directed against a second epitope on the same molecule, for 90 min at room temperature. Following the incubation, unbound conjugate was removed by washing and the bead was incubated with o-phenylenediamine solution for 30 min at room temperature. By using a Dynatech MR700 Microelisa reader (Dynatech Laboratories Inc, Chantilly, VA) set at 492 nm, the absorbance of each sample was measured. Since the amount of serum IL-2R present is directly proportional to the absorbance of the sample, the concentration of soluble IL-2R was calculated by comparing it to already known standards containing IL-2R. The minimum detectable concentration of IL-2R was 50 U/ml. Its normal range (the mean value ±2 SD) was determined by measuring the concentrations in 97 healthy control subjects (52 men, 19 to 63 years old, and 45 women, 21 to 43 years old).

Assay for Serum Angiotensin Converting Enzyme Activity

The level of serum ACE activity was determined by the modified spectrophotometric assay of Cushman and Cheung as described previously. The values of less than 40 U/ml were considered normal.

Bronchoalveolar Lavage

Bronchoalveolar lavage fluid (BALF) was obtained within one week after serum samples were collected from nine patients with active untreated sarcoidosis. A flexible fiberoptic bronchoscope was wedged in a segmental bronchus of the right middle lobe and three 50-ml aliquots of 0.9 percent sterile saline solution were injected. Total cell number was calculated and a cytocentrifuge preparation was stained with Wright-Giemsa staining. A differential cell count was obtained by counting 500 cells, and surface markers of lymphocytes were examined by using FITC-conjugated anti leu4/PE-conjugated anti leu12 solution (CD3+/CD19+ Simultest T and B Cell Test, Becton Dickinson & Co, Oxnard, CA) and FITC-conjugated anti leu3/PE-conjugated anti leu2a solution (CD4+/CD8+ Simultest T Helper/Suppressor Test, Becton, Dickinson & Co, Oxnard, CA). Fluorescence intensity of each sample was measured by using fluorescence-activated cell sorter (FACStar, Becton, Dickinson & Co, Oxnard, CA).

Statistical Analysis

The statistical significance of differences was determined by using Student's t test. Comparison between two parameters was made by using Spearman's regression analysis.

RESULTS

Serum Concentrations of Soluble IL-2R in Active, Dormant and Corticosteroid-Treated Sarcoidosis

Serum concentrations of soluble IL-2R were compared among the three groups described previously. The mean value of soluble IL-2R concentrations in serum samples from 34 patients with active untreated sarcoidosis was 1,143 U/ml and that for 30 patients with dormant sarcoidosis was 353 U/ml, and the difference was highly significant (p<0.001; Fig 1), while the normal range was 80 to 300 U/ml in 97 healthy control subjects. The mean value for patients with treated sarcoidosis was 380 U/ml and the difference between treated and untreated patients with active disease also was significant (p<0.001).

Correlation between Serum Concentration of Soluble IL-2R and Serum Angiotensin-Converting Enzyme Levels

Figure 2A shows the relationship between serum
concentrations of soluble IL-2R and serum ACE levels in active untreated sarcoidosis, showing a positive correlation ($r = 0.76$). Serum ACE levels from six of 34 patients were within normal limits. On the other hand, values of serum-soluble IL-2R were above the normal level in all patients with active untreated sarcoidosis. The values of both parameters were low in most of the patients with dormant sarcoidosis (Fig 2B), but in one patient both of the two parameters remained significantly high after chest x-ray film and ophthalmologic examinations showed resolution of abnormalities. No obvious dissociation between these two parameters was noted in patients with untreated sarcoidosis.

**Correlation of Soluble IL-2R in Serum with Some Parameters of BAL Fluid**

The relationship between soluble IL-2R concentration and some parameters of BAL fluid was studied in nine cases with active sarcoidosis, and no apparent association was observed between serum concentration of soluble IL-2R and the percentage of lymphocytes ($r = 0.07$) or CD4+/CD8+ ratio of the lymphocytes in the BAL fluid ($r = 0.08$, figures not shown).

An attempt was made to measure the soluble IL-2R levels in the nonconcentrated cell-free lavage fluid; however, they were below the detection limit in all of these patients.

**Changes in Serum Concentrations of Soluble IL-2R during the Course of the Disease**

Changes in serum concentrations of soluble IL-2R and ACE activity were monitored in some patients with sarcoidosis. The following are representative cases.

Figure 3A shows serial changes in concentrations of serum-soluble IL-2R and serum ACE activities in a patient with spontaneous remission of the disease. The patient, a 56-year-old man, was diagnosed as having sarcoidosis with uveitis and BHL in October 1986. Levels of both parameters were elevated at the time of diagnosis, and they decreased to the normal range in six months along with improvement of eye symptoms and resolution of chest abnormalities evidenced radiologically.

The second patient was a 55-year-old woman who developed neurologic and eye symptoms due to sarcoidosis. Corticosteroid therapy was begun with an...
initial dosage of 30 mg of prednisolone per day, and serum concentration of soluble IL-2R decreased rapidly to the normal range within four weeks, accompanied by improvement of her symptoms and a decrease in serum ACE activity (Fig 3B). A similar rapid decrease in both the serum soluble IL-2R level and ACE activity also was noted in other patients who received corticosteroid treatment (data not shown).

The third case was a 33-year-old man who manifested the disease with uveitis and BHL. Corticosteroid therapy had been started at another hospital (initial dosage of 30 mg of prednisolone daily), which was continued for eight months; resolution of eye symptoms and chest abnormalities evidenced radiologically ensued. After cessation of the therapy, reappearance of chest abnormalities evidenced radiologically and elevation of serum concentration of soluble IL-2R and ACE activity were noted (Fig 3C).

**DISCUSSION**

In the present study, we confirmed that serum concentrations of soluble IL-2R were elevated in untreated patients with active sarcoidosis as compared with healthy controls. The level in untreated active sarcoidosis was significantly higher than in those with dormant or treated sarcoidosis. In patients with spontaneous remission, the level gradually fell to normal range along with the resolution of chest abnormalities evidenced radiologically. In corticosteroid-treated patients, it rapidly decreased to normal range within four weeks after initiation of the therapy. We also observed a correlation between serum concentration of soluble IL-2R and serum ACE activity, and the association noted in our study was stronger than that reported previously. Serial measurements of those two parameters in patients with spontaneous remission or those treated with steroids showed that the change in serum IL-2R level well paralleled that in serum ACE activities. These results indicate that serum IL-2R level could be used as a marker of disease activity in sarcoidosis.

In terms of sensitivity of these two parameters in detecting disease activity of sarcoidosis, we found less overlap of values between active sarcoidosis and healthy control subjects in serum-soluble IL-2R levels than in serum ACE activity. Determining IL-2R concentrations might be more laborious and expensive than measuring ACE activity. The former appears to be more useful in discriminating active from dormant patients in sarcoidosis patients.

Although most cases showed a good correlation between serum IL-2R levels and clinical or chest radiologic findings, one of our cases with clinically inactive sarcoidosis showed significantly high levels of both parameters of serum IL-2R concentration and ACE activity. By assessment with BAL findings, Wallaert and co-workers observed lymphocytic alveolitis with granulomatous infiltration of the lung in their patients with extrathoracic sarcoidosis without any evidence of thoracic involvement of the disease estimated by clinical, functional or radiologic studies. These results suggest that conventional chest x-ray film studies or pulmonary function tests may not be sensitive enough to detect slight pulmonary involvement in sarcoidosis. Elevated levels of both serum IL-2R and ACE activity of one of our patients with dormant sarcoidosis described previously might be due to some granulomatous lesions that were not manifested by clinical examinations or chest x-ray film...
There were no obvious association between the level of serum concentration of soluble IL-2R and the percentage of lymphocyte or CD4+/CD8+ ratio in our preliminary study. Lawrence et al. found a significant increase in the soluble IL-2R level in the concentrated lavage fluid in two thirds of their patients with sarcoidosis. Although measuring soluble IL-2R level in the BAL fluid is not as clinically useful as that in serum samples because of the low level of soluble IL-2R in the BAL fluid and the overlap with data between patients with sarcoidosis and the normal group, investigations in this field seem to be important to clarify the relative roles of the lung in the increased level of serum soluble IL-2R in sarcoidosis.

We concluded that the serum concentration of soluble IL-2R is a sensitive parameter reflecting the disease activity in sarcoidosis. Further studies on the precise mechanism of this subject are needed.

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REFERENCES