Serum antigammaglobulins in juvenile rheumatoid arthritis

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In cases of juvenile rheumatoid arthritis (JRA), rheumatoid factors (RF) in the serum, when tested by the usual agglutination reactions, are found much less frequently than in cases of adult rheumatoid arthritis. The reason for this is not known. We have already reported the results of our study of RF negativity in JRA (Bardfeld and Houba, 1964) and have shown comparatively high values of the immunoglobulins IgG, IgA, and IgM (Houba and Bardfeld, 1969).

The purpose of the present work was to detect levels of serum antigammaglobulins (AGG) using a new technique and to compare them with the levels of IgG, IgA, and IgM in the sera of patients with JRA.

Material and methods

Sera were obtained from 32 patients with JRA all fulfilling accepted diagnostic criteria (Ansell and Bywaters, 1963). The age range was from 3 to 20 years (mean 13). Sera were also taken from 42 healthy young subjects aged from 3 to 17 years (mean 6).

AGG were determined by our own quantitative method. Nitrocellulose membrane ultrafilters (Synpor 6, VCHZ-Synthesia, Pardubice, Czechoslovakia) are impregnated for 10 minutes with a 1 per cent. solution of human gammaglobulin (Cohn fraction II). After washing away excess protein (three washings in saline pH 7.4 for 10 minutes under constant shaking are satisfactory), 5 μ l. of the tested serum are spotted on the membrane. After the serum is absorbed, unbound proteins are washed away from the ultrafilter (four washings with saline pH 7.4 for 10 minutes remove all soluble proteins). The membrane is then stained with buffered amidoblack 10B solution (0.1 per cent. solution in 0.2M sodium acetate pH 4.6). The excess stain is removed by three washings for 5 minutes in a 3 per cent. solution of acetic acid. The intensity of the stain of each spot is measured by spectrophotometer Spekol equipped with a reflectometer attachment at the wavelength 580 μ m. The results are read on the calibration curve obtained by using known amounts of human immunoglobulin IgM (Soběslavský and Hladíková, 1968).

Immunoglobulins were determined by the technique of Fahey and McKelvey (1965), using the commercial antisera to IgG, IgA, and IgM and the related reference proteins (Institute of Sera and Vaccine, Prague, Czechoslovakia).

All sera were tested for RF by the latex-fixation and sensitized sheep cell agglutination tests (Houba and Allison, 1966; Houba and Bardfeld, 1969).

Results

The levels of AGG (mg./ml.) are shown in Fig. 1. The healthy subjects are represented by individual points in the first column, and the patients with JRA in the second. The mean value is indicated in each column by a horizontal line. In the healthy controls this value is 3.9, in the JRA patients 5.1. Among our series of 32 cases of JRA, 53 per cent. had raised values of AGG and 47 per cent. were within normal limits. A lower value of AGG was not seen in any sick subject. The difference between these two groups is significant ($P < 0.01$).

FIG. 1 Levels of antigammaglobulins in 42 normal and 32 JRA sera shown by points, mean values by horizontal lines.

The IgG, IgA, and IgM levels of JRA patients are shown in Fig. 2. Cases with raised AGG values are indicated by circles and those with normal levels, by dots. The raised values of AGG are distributed with equal frequency among all the values of the individual immunoglobulins.

FIG. 2 Serum levels of individual immunoglobulins in 32 cases of juvenile rheumatoid arthritis; seventeen with raised $\circ$, and fifteen with normal $\bullet$ values of antigammaglobulins.

Among the 32 patients with JRA, rheumatoid factors were detected by the latex-fixation technique

in two cases, and in one of them, in addition, by the sensitized sheep cell agglutination test. The sensitized sheep cell agglutination test was positive in one of the 42 healthy subjects.

Our findings of raised values of AGG in cases of JRA correspond with data recently reported by other authors. Kss and Munthe (1969) described the same frequency of findings of 'pepsin-agglutinators' by using haemagglutination reactions. On the other hand, Torrigiani, Ansell, Chown, and Roitt (1969) found raised values of IgG antiglobulins in all active cases of JRA by the quantitative immunoadsorption technique. This statement also confirms the opinion that JRA patients are still serologically active even when RF are not detectable by latex-fixation and/or sensitized sheep cell agglutination tests.

The behaviour of AGG in other diseases is to be the subject of a further study.

Summary

Raised levels of antigammaglobulins were found in the sera of seventeen out of 32 patients with juvenile rheumatoid arthritis, compared with 42 healthy young subjects. The distribution of raised values of antigammaglobulins in patients with juvenile rheumatoid arthritis was independent of the values for individual immunoglobulins (IgG, IgA, and IgM).

The authors wish to thank Dr. B. M. Ansell and Dr. A. Weston for help in the preparation of the manuscript.

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