

Significance of Serologic Abnormalities in Lepromatous Leprosy¹

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Patients with lepromatous leprosy show a wide range of seriological abnormalities including variations in the levels of serum immunoglobulins (4, 17, 21), depression of cell-mediated immunity (3, 18, 20), and the presence of several types of autoantibodies (14, 19). Occurrence of these phenomena in a disease of known infectious etiology provides an excellent opportunity to study the interrelationship and the significance of such abnormalities. In an attempt to see if there is any correlation between the abnormalities of serum immunoglobulins, depression of cell-mediated immunity and the presence of autoantibodies, we have estimated the levels of immunoglobulins (IgG, IgM, IgA), rheumatoid factor (RF), antithyroid antibody (ATAB), antinuclear antibody (ANAB) and the hepatitis-associated-antigen (HAA) in patients having lepromatous leprosy and compared the results with similar investigations undertaken on apparently healthy individuals and patients suffering from so-called autoimmune diseases. Serum levels of complement (C₃) and C-reactive protein (CRP) have also been studied.

MATERIALS AND METHODS

Serum samples were obtained from 50 patients having lepromatous leprosy without *erythema nodosum leprosum*, 54 normal controls taken from among the members of faculty, medical students and laboratory staff and 113 patients suffering from autoimmune diseases such as rheumatoid arthritis, thyroiditis, systemic lupus erythematosus and other collagen vascular diseases.

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The following techniques were used for detecting the serological abnormalities, though all procedures were not necessarily applied to all serum samples.

Single radial immunodiffusion technic of Fahey and McKelvey (5) for estimation of levels of immunoglobulins (IgG, IgM, IgA) and complement (C₃). The monospecific antisera used in this study were developed in our laboratory (10, 12).

Latex fixation method using RF test kit (Sylvania, USA) for the rheumatoid factor.

Passive hemagglutination method using thyroglobulin antibody test kit (Burroughs, Wellcome) for antithyroid antibody.

Horse-radish peroxidase conjugated antibody technic of Nakane and Pierce (15) as modified by Petts and Roitt (16) for antinuclear antibody.

Electro-osmo-diffusion in agar (6) for hepatitis-associated-antigen. Latex fixation method using C-R test kit (Sylvania, USA) for C-reactive protein.

RESULTS

Rheumatoid factor was detected in 26%, antithyroid antibody in 16%, antinuclear antibody in 26%, and C-reactive protein in 24% of lepromatous patients as compared to levels of 19.3%, 17.7%, 34.3% and 20.9% respectively, in samples from diseases of autoimmune group. They were absent in the sera of normal controls (Table 1).

Hepatitis-associated antigen was discovered in 14% cases of lepromatous leprosy as compared to 0.1% in normal persons as previously reported (13) from a study of 952 residents of New Delhi.

The mean serum levels of immunoglobulins and complement (C₃) in lepromatous patients showed some variations from the normal values of healthy Indians reported earlier (11), but statistically the differences were not significant (Table 2). The values of immunoglobulins have been expressed in WHO units (8) while those of complement (C₃) are given in percentages, taking the value of a pooled sample of serum from 50 apparently healthy Indian subjects as 100% (11).

TABLE 1. Comparative occurrence of autoantibodies, C-reactive protein and hepatitis-associated antigen.

| | Occurrence of abnormal findings in | | | | | | Autoimmune diseases | | |
|------------------------------|------------------------------------|--------------|------------------|---------------------|--------------|------------------|---------------------|----------|----------|
| | Normal controls | | | Lepromatous leprosy | | | No. | No. | Percent |
| | No. tested | No. positive | Percent positive | No. tested | No. positive | Percent positive | tested | positive | positive |
| Rheumatoid factor | 54/0 | 0 | | 50/13 | 26 | | 98/18 | | 19.3 |
| Antithyroid antibody | 54/0 | 0 | | 50/8 | 16 | | 113/20 | | 17.7 |
| Antinuclear antibody | 50/0 | 0 | | 50/13 | 26 | | 67/23 | | 34.3 |
| C-reactive protein | 54/0 | 0 | | 50/12 | 24 | | 67/14 | | 29.9 |
| Hepatitis-associated antigen | 952/1 | 0.1 | | 50/7 | 14 | | — | | — |

TABLE 2. Levels of immunoglobulins and complement (C₃) in normal controls and lepromatous leprosy patients.

| Serum protein | Normal controls | Lepromatous leprosy | p value |
|----------------|-------------------------------|---------------------|---------|
| IgG | 172 (120-246) ^a | 299 (151-591) | > 0.4 |
| IgM | 175 (108-284) | 319 (58-1750) | > 0.1 |
| IgA | 121 (55-268) | 164 (35-761) | < 0.02 |
| C ₃ | 101 (62-163) | 82 (28-236) | < 0.02 |

^a Figures in parentheses indicate 2 SD range from the mean.

DISCUSSION

This study indicated that autoantibodies occur in the patients of lepromatous leprosy with almost the same frequency with which they occur in patients suffering from the autoimmune diseases. If *Mycobacterium leprae* were not known to cause this disease, it would be tempting to call lepromatous leprosy an autoimmune disease. The presence of autoantibodies must be regarded only as a paraphenomenon in view of the known etiology of leprosy. This should suggest that the mere discovery of autoantibodies in a disease of unknown etiology should not lead to a hasty conclusion that that disease is autoimmune. It is important to satisfy the criteria of Humphry and White (7) for labeling the disease to be autoimmune and demonstrate the pathogenic capacity of the autoantibody.

The cause of the development of autoantibodies in patients of lepromatous leprosy is a matter of speculation. Türk and Bryceson (18) have suggested that *Mycobacterium leprae* may be acting as an adjuvant in the immunization of the patient to his own antigens. It has also been postulated that the natural tolerance to self-antigens can be broken by an infectious agent if the latter shares some antigenic determinants with the self-antigens. This, however, seems to be unlikely in this case because it is highly improbable that *Mycobacterium leprae* has antigenic determinants common with all the antigens against which these autoantibodies are directed.

Recently Allison *et al* (1), advanced the hypothesis that thymus-dependent T-lymphocytes play a key role in preventing autoimmunity. It is thus probable that dis-

eases associated with a depressed state of cell-mediated immunity may be more prone to develop autoantibodies. Several recent studies (3, 18, 20) have provided evidence that a significant percentage of patients of lepromatous leprosy are associated with a marked depression of cell-mediated immunity as manifested by failure to develop contact hypersensitivity, defective lymphoblast transformation and delayed rejection of homograft. Chronic persistence of hepatitis-associated antigen has also been associated with a depression of cell-mediated immunity (2, 9). The high incidence (14%) of hepatitis-associated antigen in leprosy patients provides further evidence of the depression of cell-mediated immunity in lepromatous leprosy. To check the above hypothesis of Allison *et al*, an attempt was made to see if autoantibodies occur more frequently in patients showing persistence of hepatitis-associated antigen in their sera. Table 3 shows that autoantibodies occur as frequently in sera of patients with hepatitis-associated antigen as without it.

Sheagren *et al* (17), and Bullock *et al* (4) reported a rise in IgG and IgA in lepromatous leprosy, while Waters *et al* (21) reported an increase in IgA and IgM. In our study the mean values of the major immunoglobulin classes were higher in leprosy patients than our normal controls. But the log method of statistical analysis (11), however, showed that these differences were not significant. Such statistical analysis is necessary in this instance because of the skewed deviation found in the distribution of immunoglobulin levels in normal persons. Nevertheless some patients did have very high levels of IgG or IgM, but those showing high levels of IgM were not necessarily those having the rheuma-

toid factor in their serum and patients showing raised IgG levels were not the ones having antinuclear and/or antithyroid antibodies. This suggests that the rise in the levels of immunoglobulins is due to many factors, one of which may be the presence of autoantibodies.

Serum levels of complement (C₃) have been reported to be high in lepromatous patients but only with *erythema nodosum leprosum* (ENL) (21, 22). We did not find any significant difference from our normals and none of our patients had ENL. The levels of C₃ in different patients followed the same range irrespective of whether the patient had CRP or not.

SUMMARY

Patients having lepromatous leprosy but without ENL, showed the presence of autoantibodies such as rheumatoid factor, antithyroid antibody and antinuclear antibody and C-reactive protein with almost the same high frequency as patients suffering from the so-called autoimmune diseases. It is stressed, however, that the presence of autoantibodies in leprosy is only a paraphenomenon and does not indicate that leprosy is an autoimmune disease. Hepatitis-associated antigen was present in 14% of serum samples from leprosy compared to 0.1% in normal individuals. There was no association between the chronic persistence of hepatitis-associated antibody and the presence of autoantibodies. The levels of immunoglobulins IgG, IgM and IgA, and complement (C₃) in patients with lepromatous leprosy showed wide variations, but the differences from normal controls were statistically not significant. There was no correlation between the raised level of immunoglobulins in leprosy

TABLE 3. Persistence of hepatitis-associated antigen with various autoantibodies in lepromatous leprosy.

| Hepatitis-associated-antibody | ANAB | | RF | | ATAB | |
|-------------------------------|----------|----------|----------|----------|----------|----------|
| | Positive | Negative | Positive | Negative | Positive | Negative |
| Positive (7 cases) | 2 | 5 | 3 | 4 | 1 | 6 |
| Negative (43 cases) | 11 | 32 | 10 | 33 | 7 | 36 |
| Total | 13 | 37 | 13 | 37 | 8 | 42 |

patients and the presence of autoantibodies.

RESUMEN

Se demostró, en pacientes con lepra lepromatosa pero sin ENL, la presencia de autoanticuerpos tales como factor reumatoideo, anticuerpos anti-tiroglobulina, anticuerpos antinucleares y proteína C-reactiva, con una frecuencia casi igualmente alta que en pacientes que sufrían de las así llamadas "enfermedades auto-inmunes". Se desea recalcar, sin embargo, que la presencia de auto-anticuerpos en lepra es sólo un parafenómeno y no indica que la lepra sea una enfermedad auto-inmune. El anticuerpo asociado con hepatitis se encontró en 14% de los sueros de pacientes con lepra, en comparación con un 0,1% de los sueros de individuos normales. No se encontró asociación entre la persistencia crónica del anticuerpo asociado con la hepatitis y la presencia de auto-anticuerpos. Los pacientes con lepra lepromatosa mostraron amplias variaciones en sus niveles de inmunoglobulinas IgG, IgM e IgA y complemento (C₃), pero la diferencia con los controles normales no fué estadísticamente significativa. No se observó relación entre el aumento del nivel de inmunoglobulinas y la presencia de auto-anticuerpos en los pacientes con lepra.

RÉSUMÉ

Des malades atteints de lèpre lépromateuse, mais ne souffrant pas d'érythème noueux lépreux (ENL), ont révélé la présence d'auto-anticorps tels que le facteur rhumatoïde, des anticorps anti-thyroïdiens, l'anticorps anti-nucléaire, et la protéine C-réactive, avec presque la même fréquence que des malades souffrant des maladies dites auto-immunes. On souligne cependant que la présence d'auto-anticorps dans la lèpre n'est qu'un phénomène marginal, et qu'elle n'indique pas que cette maladie est une affection auto-immune. L'antigène associé à l'hépatite (HAA) était présent dans 14 pour cent des échantillons de sérum provenant de malades de la lèpre, mais dans 0,1 pour cent seulement des sérums provenant d'individus normaux. On n'a pas constaté d'association entre la persistance chronique de l'anticorps associé à l'hépatite (HAA) et la présence d'auto-anticorps. Le taux des immunoglobulines IgG, IgM et IgA, de même que le taux du complément (C₃), chez des malades souffrant de lèpre lépromateuse, ont présenté de larges variations, mais les différences par rapport aux témoins normaux étaient sans signification statistique. Il n'y avait pas de corrélation entre l'élévation du taux des

immunoglobulines chez les malades de la lèpre, et la présence d'auto-anticorps.

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