

Fluorescent antibody study for antinuclear antibodies in bronchial asthma

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Fluorescent antinuclear antibody studies and LE cell tests for antinuclear antibodies were completely negative in a series of cases of chronic bronchial asthma. Therefore, it is concluded that the formation of antinuclear antibodies, as found in collagen diseases, does not occur in bronchial asthma.

Studies have been performed to determine whether antinuclear antibodies occur in chronic bronchial asthma like those found in collagen diseases such as disseminated lupus erythematosus and rheumatoid arthritis. A number of authors have suggested that collagen diseases are manifestations of an allergic reaction.^{1, 2} If antinuclear antibodies could be demonstrated in cases of bronchial asthma, this would be evidence of a characteristic common to an allergic disease and collagen diseases.

The lupus erythematosus (LE) cell phenomenon is based on the presence of an antinuclear antibody. The LE factor is a 7S gamma globulin and it has the properties of an antibody against cell nuclei or, more specifically, against nucleoproteins. The antibody reacts with desoxyribonucleic acid (DNA) of various sources (bovine, thymus, bacteriophage, etc.).³ In a previous paper⁴ we reported that LE cell tests were completely negative in a series of cases of severe chronic bronchial asthma in which complete studies were performed to exclude coexisting collagen diseases. This was evidence against the occurrence of antinuclear antibodies in bronchial asthma.

Dr. Paul Calabresi and his associates⁵ studied cases of ulcerative colitis for evidence of the LE cell phenomenon and circulating antinuclear antibodies, as

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the disease has been considered possibly allergic in origin. They obtained completely negative LE cell tests but did demonstrate circulating antinuclear globulins using the fluorescent antibody-staining technique. Therefore, fluorescent antibody studies were required definitely to determine the absence of antinuclear antibodies in bronchial asthma.

A series of 11 cases of chronic bronchial asthma were studied. Ten allergic patients with ragweed hay fever, free of asthma, were used as controls. Serum was obtained from each patient.

Human gamma globulin was injected into rabbits to develop a rabbit anti-human gamma globulin antibody which was then combined with a fluorescein dye. The procedure utilized the fluorescent rabbit antihuman gamma globulin antibody to detect circulating gamma globulin with an affinity for mouse liver nuclei (indirect Coons' technique). It was performed as follows:

1. Slide imprints made from mouse liver cells were air dried.
2. Imprints were incubated with the unknown diluted serum for 30 minutes.
3. Imprints were washed 5 times.
4. Imprints were then incubated with fluorescein-labeled rabbit antihuman gamma globulin and were carefully washed with buffer.
5. A known positive LE serum was included in each test as a control.
6. All slides were examined under the fluorescent microscope.

If there were any antinuclear antibodies present in the unknown sera or sera from asthmatic patients, they would become attached to the nuclei of the mouse liver cells. The rabbit antihuman gamma globulin antibody carrying the fluorescent dye would in turn become attached to the human antinuclear antibodies previously linked to the mouse liver nuclei, thereby producing strong fluorescence of the mouse nuclei.

The known positive LE control sera caused strong fluorescence of mouse nuclei in all tests, showing the presence of antinuclear antibodies.

The 21 unknown sera, 11 from cases of asthma and 10 sera from controls with hay fever, failed to produce fluorescence and all were negative. Therefore, no antinuclear antibodies were demonstrated in cases of bronchial asthma with the use of this fluorescent antinuclear antibody technique.

In vitro LE cell tests were also performed in these cases of bronchial asthma and were also negative.

SUMMARY

The results of the previous pilot study for LE cell phenomenon in bronchial asthma were all negative. The results in this study for antinuclear antibodies in bronchial asthma with the fluorescent antibody technique and LE cell determination were negative. If a substantial number of positive tests had been obtained, a larger study would have been indicated. If a few positives had been found, one could suggest that this might be the normal incidence of antinuclear antibodies. Actually the percentage of antinuclear antibodies in apparently healthy persons was reported to be 4 per cent by Alexander and his associates,⁶ and 2 per cent by Seligmann and his co-workers.⁷ U. Serafini and his colleagues⁸ reported only 13 positive cases in 1,430 normal subjects, 0.9 per cent. But ac-

tually all tests were completely negative which is certainly no more than can be found in normal subjects.

The exact mechanism responsible for the production of antinuclear antibodies is still unknown. There may be special autoantigens formed in collagen diseases to cause the production of antinuclear antibodies, or these antibodies may be the result of an aberrant function of the immune mechanism. In view of the absence of antinuclear antibodies in these studies, it is apparent that neither of these phenomena occurs in bronchial asthma.

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