

# ANTIGENIC STRUCTURE OF THE SPLEEN IN THE POSTNATAL DEVELOPMENT OF LINE-A MICE

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Numerous experiments [3-5, 7] have shown that in most animals a state of immunologic tolerance can be created not only in the period of their embryonic development, but also for some time during their post-natal life (the "adaptive period").

Several investigators [1, 2] have shown that in the process of embryonic development, at each stage of ontogenesis antigens specific for that particular stage appear in the developing tissues. Later these are replaced by new, corresponding to the changing character of biosynthesis from stage to stage. On the basis of these facts the authors have postulated that the development of immunologic competence of the cells is associated with a modification of their antigenic properties.

In the present investigation the antigenic structure of the spleen was studied in the course of the post-natal development of mice.

## EXPERIMENTAL METHOD

Antisera were obtained against the spleen of line A mice aged 1, 7, 30, and 90 days by immunizing rabbits with saline extracts (1:10) and homogenates of that organ. Chinchilla rabbits weighing 2.0-2.5 kg were immunized three times each week for a month. The antisera thus obtained, with titers of about 1:3500, were tested in the micromodification of Ouchterlony's gel-precipitation reaction. To obtain antisera of narrow specificity, they were absorbed by Björklund's method [6]. The protein content of the antigens was determined by Lowry's method and equalized.

The reaction was set up in 1% Difco agar in 0.85% physiological saline. The antiserum for testing was poured into the central wells and the antigens from the spleen tissues into the peripheral wells. The control consisted of antigens from the tissues of a spontaneous mammary gland adenocarcinoma from line A mice and of the spleen from line C57BL mice aged 1 and 90 days. The reaction was read after 48 h.

## EXPERIMENTAL RESULTS

The antiserum against the spleen of day old mice was first tested with antigens from the spleen of mice of different ages. As is clear from Fig. 1, A this antiserum formed one precipitation line more with the antigens from the spleen of mice 15, 30, and 90 days old. After absorption of this antiserum with spleen antigens of day old mice complete exhaustion was produced, and it did not react with any of the antigens. Absorption of the antiserum with spleen antigens of 7-day mice left an ill defined precipitation line with the spleen antigens of the day old mice (Fig. 1, B). However, after absorption of the antiserum with spleen antigens from the older mice, one precipitation line remained with the spleen antigens of the mice aged 1 and 7 days (Fig. 1, C). Hence it may be concluded that the spleen of mice aged 1 and 7 days contains antigens which are absent from the spleen of the older mice.

The antiserum against the spleen of mice 30 days old was investigated in the same way. It is clear from Fig. 2, A that this antiserum formed different numbers of precipitation lines with the spleen antigens from mice of different ages. After its absorption by spleen antigens of day old mice (Fig. 2, B) and of mice 7 days old (Fig. 2, C), precipitation lines with spleen antigens of the older mice remained. Absorption by spleen antigens of the mice aged 30 and 90 days completely exhausted the antiserum, and it formed no precipitation lines with any of the antigens.

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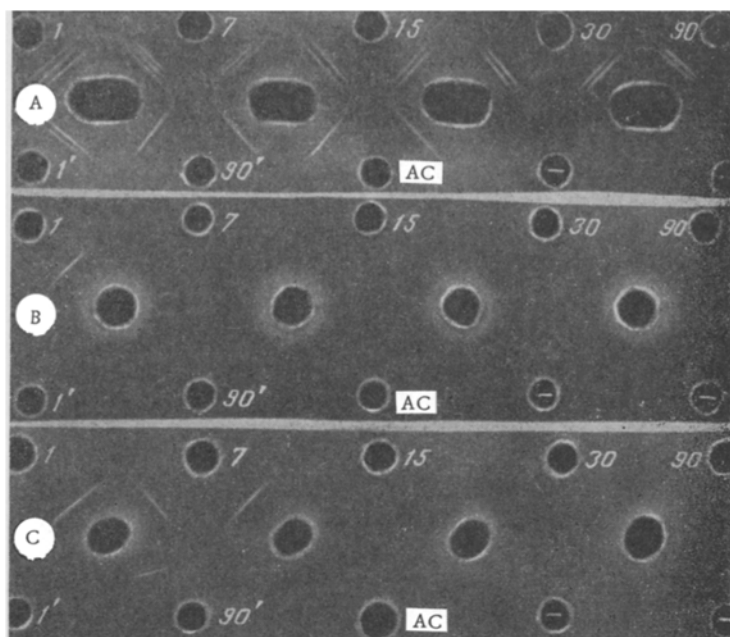


Fig. 1. Gel-precipitation reaction between antiserum against spleen of day old animals and spleen antigens of mice of different ages. A) Untreated antiserum; B) antiserum absorbed by spleen antigens of animals 7 days old; C) antiserum absorbed by spleen antigens of mice ages 15, 30, and 90 days. 1,7,15, 30,90) antigens from the spleen of line A mice of the corresponding ages, 1', 90') antigens from the spleen of line C57BL mice of the corresponding ages; AC) antigens from tissues of a spontaneous adenocarcinoma of the mammary glands of line A mice.

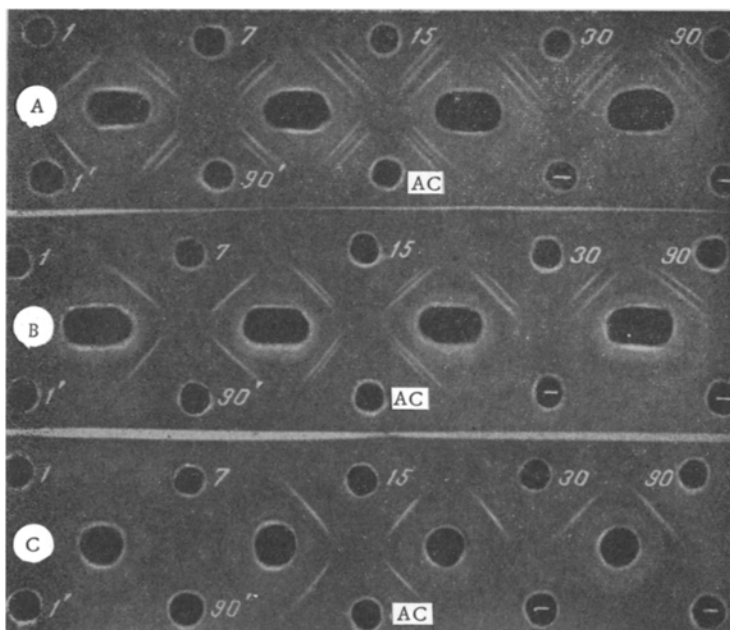


Fig. 2. Gel-precipitation reaction between antiserum against spleen of mice aged 30 days and spleen antigens of mice of different ages. A) Untreated antiserum; B) antiserum absorbed by spleen antigens of day old animals; C) antiserum absorbed by spleen antigens of mice aged 7 days. Remainder of legend as in Fig. 1.

Three differences in the antigenic structure of the spleen of newborn and mature mice cannot be explained entirely by the serum proteins contained in the tissue extracts, for when crossed gel-precipitation tests were carried out with antiserum against the spleen of day old animals, the precipitation lines formed by the plasma of the newborn mice and the spleen antigens of the mice aged 1 and 30 days were not identical. After absorption of the antiserum by newborn plasma, complete exhaustion was not observed, for it continued to form two bands with the spleen antigens of the day-old animals and one with spleen antigens of mice aged 30 days. Similar results were obtained when antiserum against the spleen of the mice 30 days old was exhausted by the plasma of newborn and mature animals.

It may be concluded from the observations described above that in the course of the postnatal development of the organism the antigenic structure of the spleen of line A mice changes. The antigenic differences between the spleen tissues at different periods of postnatal development demonstrate changes in the

metabolism of the spleen at these times. Since the times of these changes coincide with the adaptive period during reproduction of the tolerance phenomenon, it may be postulated that these changes reflect certain special features of the metabolic processes taking place in the spleen in the adaptive period.

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