AUTOANTIBODIES TO THYMUS AND SKIN EPITHELIUM IN NZB/N MICE AND (NZB × NZW)F_1 HYBRIDS

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It has now been shown that the epithelium of the mammalian thymus constitutes a microenvironment where T lymphocytes can mature and differentiate [4]. The basis of this microenvironment is formed by the "nurse cells" of the thymus epithelium, which contain Ia antigens on their surface [10]. In NZB mice and (NZB × NZW)F_1 hybrids (B/W), atrophy of the thymus epithelium is known to take place before the beginning of the autoimmune process (in the first month of life) [9]. It has also been shown that the blood thymulin level falls during the first few months after birth in NZB mice and the content of the special population of thymus epithelial cells producing this hormone decreases [7]. When thymus epithelial cells of New Zealand mice are grown in culture, they degenerate rapidly and undergo vacuolation. According to the authors cited, this phenomenon is due to a genetically programmed defect of the epithelial tissue, which is the main cause of disturbance of function of the sinus and development of the autoimmune process in NZB and B/W mice [3]. One basis of this hypothesis is the restoration of some immunologic functions of the thymus, such as the suppressor function of T lymphocytes, when B/W lymphocytes are subjected to the action of normal thymus epithelial cells of BALB/c mice [3].

It has been suggested that disturbance of the maturation of suppressor T cells and the consequent appearance of cellular autoimmune responses characterisic of the development of autoimmune processes are due to injury to the thymus epithelium by autoantibodies [2, 6].

Accordingly the aim of the present investigation was to look for autoantibodies reacting with thymus epithelial cells in NZB3N mice and B/W hybrids during the first month of life.

Data showing the ectodermal origin of the epithelial cells of the thymus were obtained previously [1, 5, 6]. Accordingly, when looking for autoantibodies to thymus epithelium, we investigated sections of thymus and skin tissues simultaneously.

EXPERIMENTAL METHODS

Blood sera obtained in the 1st-2nd and 4th-5th months after birth from NZB/N and B/W mice, and also from control mice (BALB/c) were tested. The investigations were carried out by the indirect immunofluorescence method [6] on unfixed frozen sections of thymus tissue from mice of the above-mentioned lines, guinea pigs, and man (thymus tissue of 18-week human fetuses and from adults dying from trauma, blood group 0). Sections of mouse and guinea pig skin (from the lip) and human lip (skin from the region of a joint from 18-week fetuses) were used. Some investigations were conducted in syngeneic and autologous systems (on tissues of NZB/N or B/W mice from which sera were obtained). To inhibit reactions of the test sera with cell nuclei, we used native DNA obtained from calf thymus (2 mg DNA to 0.2 ml serum in a dilution of 1:8). The preparations were examined under the ML-2 luminescence microscope with 40 × objective; the homal 3 was used for photography.

EXPERIMENTAL RESULTS

Sera from NZB/N mice aged 1-2 months, when tested on skin sections from BALB/c mice reacted in 11 of 13 cases with the stratum basale of the skin epithelium (Fig. 1a). Sera ob-

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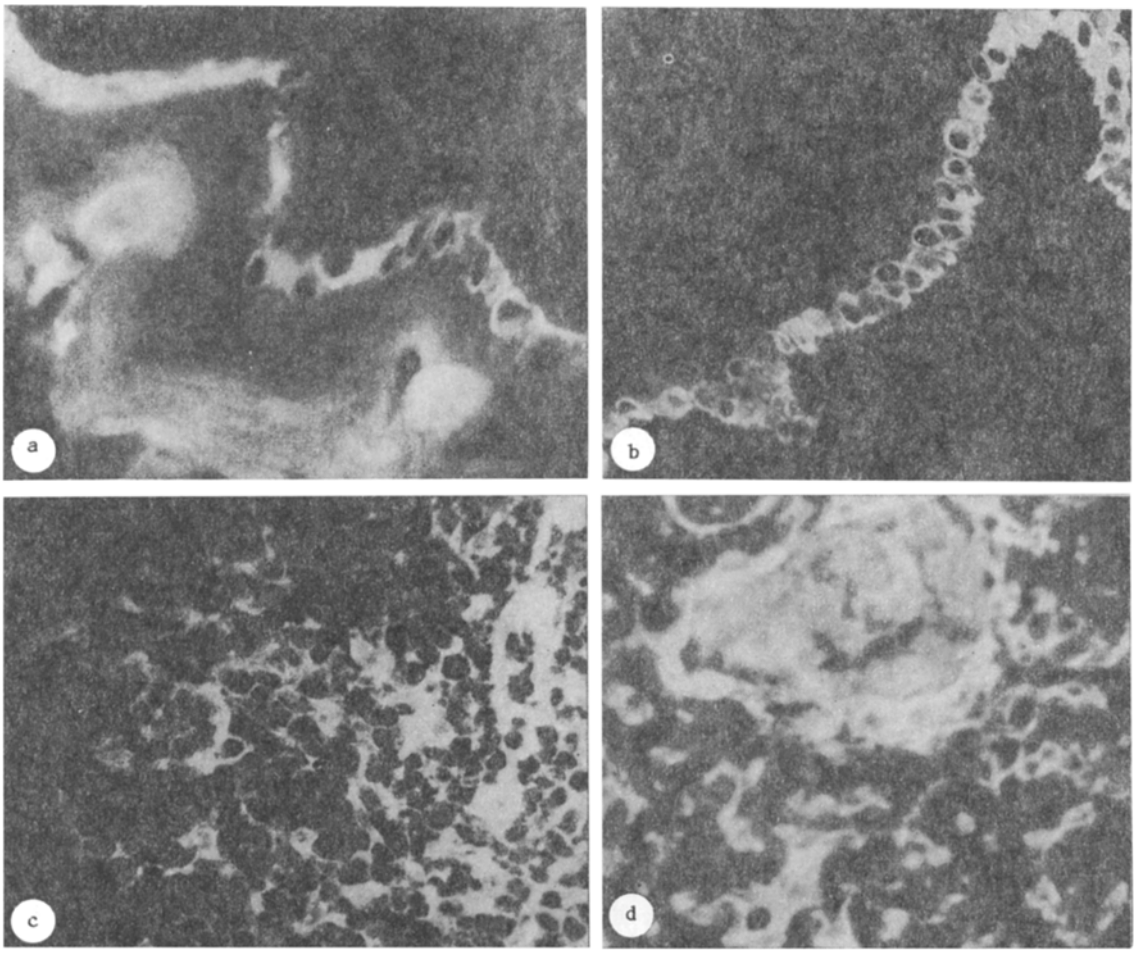

Fig. 1. Sera from NZB/N mice aged 1-2 months tested by the indirect immunofluorescence method: a) reaction with stratum basale of skin epithelium of BALB/c mouse; b) rection with stratum basale of human skin epithelium; c) reaction with epithelium of cortical zone of thymus in autologous system; d) reaction with epithelial cells in medullary zone around Hassall's corpuscles in section through human thymus.

tained from B/W mice of the same ages also reacted in 13 of 17 cases with the above-mentioned structures. Later (age 4-5 months) reactions with the stratum basale of the skin epithelium were observed when all sera of NZB/N (eight sera) and B/W mice (13 sera) were tested. Reactions were observed in dilutions of the sera from 1:8 to 1:128. None of the 23 sera from BALB/c mice, taken at the same ages, reacted when tested on the stratum basale of the skin epithelium in dilutions of 1:8 or more. Positively reacting sera from NZB/N and B/W mice were subjected to parallel tests on skin sections in autologous, syngeneic, and heterologous (human and guinea pig skin) systems (Fig. 1b). In all cases fluorescence of the stratum basale of the epithelium was observed.

Further tests were carried out on sections of thymus tissues. Positive reactions with thymus epithelium were found in all the above-mentioned systems as a result of testing of sera reacting positively with skin epithelium, including syngeneic and autologous systems (Fig. 1c, d). Positive reactions with the epithelium in sections of thymus tissues from mice of all lines tested in autologous and syngeneic systems, in sections through the thymus of BALB/c mice, and also in human thymus tissues were found in the medullary and cortical zones of the thymus. In sections of the guinea pig thymus the sera reacted with the epithelium only in the medullary zone. In control experiments with sera of BALB/c mice negative results were obtained on sections of the human and guine pig thymus and the thymus from mice of the different lines.

Inhibition experiments showed that calf thymus DNA inhibits reactions with cell nuclei, observed when certain sera from NZB/N and B/W mice were tested on skin sections. Meanwhile reactions of these sera with skin and thymus epithelium were completely preserved.

Thus the presence of antibodies reacting with epithelium of the cortex and medullary zones of the thymus and also with epithelium of the stratum basale of the skin was established in NZB/N and B/W mice during the first months after birth. No such antibodies were found in BALB/c mice. The antibodies found in NZB/N and B/W mice must be taken as autoantibodies. First, positive reactions were found in autologous and syngeneic systems. Second, the antigen with which these antibodies reacted was present in all samples of thymus and skin tissues tested in man, guinea pigs, and mice of the various lines. This last fact is evidence that this antigen is a tissue-specific antigen common to different species, against which autoantibodies usually arise [2].

It will be noted that autoantibodies reacting with thymus and spleen epithelium differ from autoantibodies that react with cell nuclei. The next step must be to compare antibodies to thymus epithelium with antibodies to thymocytes, which are usually found in NZV and B/W mice [8]. It will also be interesting to look for autoantibodies to epithelium in New Zealand mice of different ages and, in particular, in newborn mice, in order to solve the problem of transmission of these autoantibodies from mother to fetus.

It must be emphasized that autoantibodies reacting with thymus epithelium were found before the beginning of the autoimmune process which, as we know, begins in NZB and B/W in the 4th month of life. The factors causing the appearance of autoantibodies to thymus epithelium and the questions of to which epithelial tissue antigens they are directed, and the role they play in the development of the autoimmune process all require further study. The role of these autoantibodies in damage to thymus epithelium and in the onset of immunoregulatory disturbances characteristic of the autoimmune process must be exlained.

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