we bear in mind the tendency of phenobarbital to lower the liver's resistance to ischemia and its known sedative effect.

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Localization and Fate of Thymic Epitheliocytes of Endodermal Origin Synthesizing Cytokeratin 18

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A reaction in the cytoplasm and processes of some cells of the epithelial reticulum is revealed by indirect immunofluorescence with monoclonal antibodies to cytokeratin 18 in the cortical zone of human thymus. In the medullary zone the reaction is observed in spherical elements similar in shape and size to intestinal goblet cells.

Key Words: *epithelial cells of the thymus; endodermal origin; cytokeratin 18*

Tissue structures characteristic of many different organs are described in fundamental works on the morphology of the thymus which are frequently cited [3,7,8]. The presence in the thymus of spheroid cells resembling in their tinctorial properties young muscle elements and of cells similar to intestinal mucosal epithelium goblet cells, in which mucin has been detected [8], is quite perplexing. By now myoid cells of the thymus have been studied in detail [1,6]. Experiments with polyclonal antibodies have revealed in them the synthesis of contractile proteins of the heart, skeletal muscles, and smooth muscles. Reports on thymic goblet cells as heteroorgan structures are far less numerous [8], although many authorities [2,5,6] are persuaded that heteroorgan elements of the thymus are directly involved in the formation of natural immunological tolerance to the organism's own antigens.

In the present study we examined the localization and fate of goblet cells in human thymus using monoclonal antibodies (MAB) to protein characteristic of the cytoskeleton of epitheliocytes of endodermal origin.

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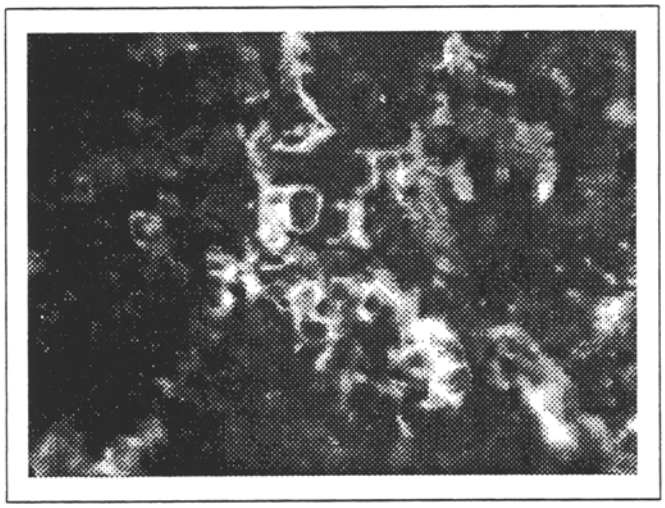

Fig. 1. Reaction in the epithelial reticulum cell cytoplasm at the border between the cortical and medullary zones (here and in other Figs. 2–4 the specimens are slices of thymus from a child aged 12).

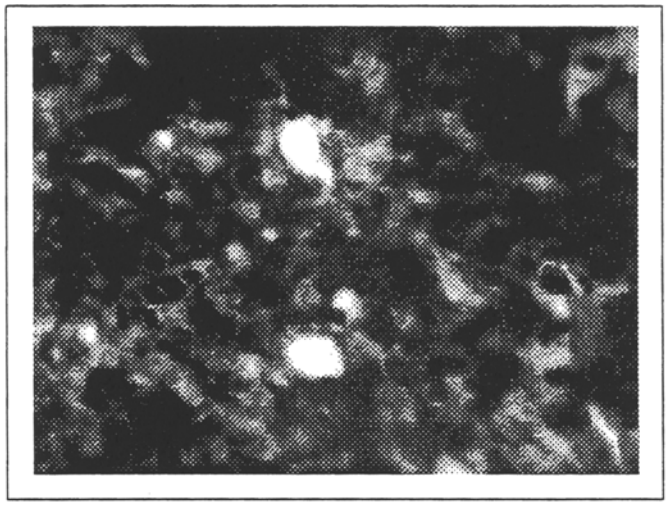

Fig. 2. Reaction in the cytoplasm of individual spherical cells resembling intestinal goblet cells (medullary zone).

MATERIALS AND METHODS

Thymic tissue from a child aged 9 who had died from acute trauma and the thymus of a 2-year-old child with congenital heart disease were used in the study. Organ tissue slices 5 μ thick were prepared in a cryostat (-20°C) and used unfixed. For indirect immunofluorescence MAB to cytokeratin 18 (Boehringer) and MAB to murine immunoglobulins labeled with fluorescein isothiocyanate (FITC) (N. F. Gamaleya Research Institute of Epidemiology and Microbiology) were used. The slices were treated with the former antibodies for 45 min and with the latter MAB for 30 min.

Slices were washed in PBS solution (pH 7.4) for 10 min and placed in neutral 60% glycerol under a slide. Corresponding controls were established.

A LYuMAM-2 fluorescent microscope was employed. RF-3 film was used to take photographs with objective 40 (water immersion) and ocular 3. Some slices were fixed in ethanol and stained with hematoxylin and eosin for histological control.

RESULTS

Examinations of thymic slices stained with hematoxylin and eosin showed a normal structure of the organ, characterized by the division of its lobules into a cortical zone rich in lymphocytes and a medullary zone with lamellar thymic (Hassall's) bodies localized against the background of far less numerous lymphocytes.

An intensive reaction in the cytoplasm of epithelial reticulum cells was detected by the indirect

Fig. 3. Cells synthesizing cytokeratin 18 join Hassall's bodies.

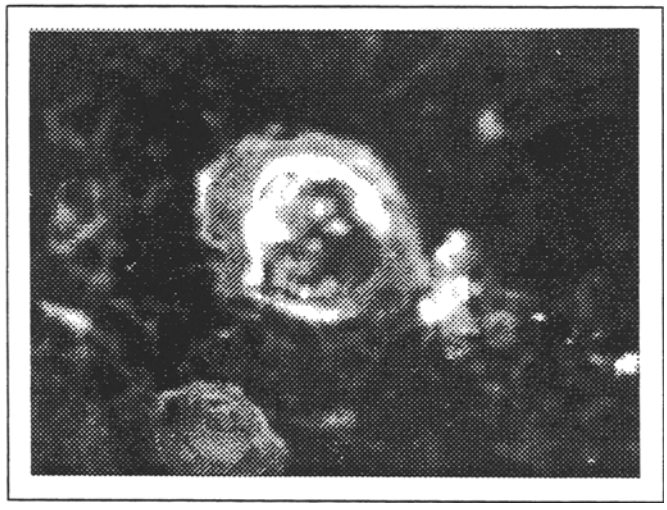

Fig. 4. Cytokeratin 18 in detritus of the central cavity of a Hassall body.

fluorescence method at the borderline between the cortical and medullary zones after treatment of thymus sections with MAB to cytokeratin 18 (Fig. 1). In the cortical zone the reaction was detectable only in individual elements or in their processes, which might be due to epithelium masking with numerous lymphocytes. In the medullary zone the reaction was localized in individual spherical or oval elements resembling intestinal goblet cells in shape (Fig. 2). Their size was not more than 20 μ, that is, smaller than medium-sized myoid cells [1]. In deeper layers of the medullary zone the cellular elements reacting with MAB to cytokeratin 18 joined Hassall's bodies, thus contributing to the formation of their outer layer (Fig. 3). A positive reaction could also be observed in the central portion of the body, which seemed to be due to death of epithelial elements and incorporation of their material in the detritus of a Hassall body cavity (Fig. 4). A similar picture has been observed in studies of the localization and fate of cellular elements synthesizing other heteroorgan antigens, notably cytokeratin of integumental epitheliocytes of ectodermal origin [5,6], muscle proteins [1,6], and lactoferrin [6]. Our observations once more confirm the results of other scientists that Hassall's bodies as structures are formed as a result of the final differentiation and death of epithelial elements of the thymus [6]. We believe that this does not preclude an active role in the physiological processes taking place in the organism of the compounds released during decay and enzymatic lysis of epitheliocyte material. This viewpoint has been expressed by many scientists [4].

Hence, cytokeratin 18, characteristic of epithelial tissues of endodermal origin, may be classed among the heteroorgan antigens of the thymus. Along with many other antigens that have now been detected in the internal medium of the thymus, this protein also appears to contribute to the formation of natural immunological tolerance to the organism's own antigens.

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