V. EXPERIMENTAL AND CLINICAL IMMUNOLOGY

MORPHOLOGICAL FEATURES OF THE RELATIONSHIPS BETWEEN EPITHELIAL CELLS AND LYMPHOCYTES IN MICE THYMUS

Ts. Marinova, E. Boshnakova

Key-words: thymus — epithelial cells — thymocytes — lymphocytes — morphology

Epithelial cells are a basic element of the so-called specific micro-environment of the thymocytes which is essential for the carrying out of the intrathymic T-lymphocytes differentiation stadium (11, 12, 23, 24). While literature data concerning the mechanisms through which epithelial cells influence lymphocytes are scanty and contradicting (6, 10, 11, 19), we took up an investigation of the morphologic relations between both these cell types in mice thymus.

Material and methods

Thymus parts of 43 conventional mice (new-born, 12-hours after birth, 7 days, 15 days, 40 days, 3 months, 4 months, 7 months) were fixed in 4 per cent glutaraldehyde (0,1 mol phosphate buffer pH—7,4) for 2 hours at 4 °C, then postfixed in 1 per cent OsO_4 (0,1 mol phosphate buffer pH—7,4) for 1 hour 30 min at 4 °C), dehydrated in ascending alcohols and embedded in Durcupan ACM. The ultrathin sections were contrasted after E. Reynolds (1963). We took the electronograms on JEM 7A.

Results and discussion

The close spatial relationships between epithelial cells and lymphocytes of mice thymus observed frequently on a large number of ultrathin sections can be divided in several basic types as follows:

1. An intimate contact between the cytolemmas of the two cells — situated in certain cell regions at a distance 30—40 nm. Sometimes we observed an invagination of the epithelial cell cytoplasm towards the neighbouring lymphocyte thus forming cytoplasmic offshoots with different shapes and size.

2. Encircling of an intact thymic lymphocyte by the cytoplasm of a neighbouring epithelial cell leading in certain areas to a sticking of their cell membranes. In some cases the encircled thymocyte shows a picnosis to a different extent.

3. A direct contact between a large part of what we called (3) "dark" cortical and medullar epithelial cells and their neighbouring lymphocytes. In the contact region under the epithelial cell cytoplasm there are elements of the rough endoplasmatic reticulum — most often dilated cisternae that are filled with a floculated, mildly electron dense material (fig. 1).

4. Contact areas between "light" thymic epithelial cells (3) and their neighbouring lymphocytes. Condensing Golgi-vacuoles (one or several), microfibrils, some of them contacting the vacuolar and/or cell membrane, coated vesicles, small Golgi-vesicles, autophagic vacuoles and solitary multivesicular bodies are concentrated in the epithelial cell cytoplasm underlying the contact.

Groups of vesicles are located not only subplasmalemmally but also in the neighbouring intercellular space. Sometimes the vacuolar membrane (of the condensing Golgi-vacuole or of the autophagic vacuole) is in close or direct contact with the cell membrane. Elements of an well-developed RER are also seen in this area. (fig. 2, a, b, c, d, e).

Fig. 1. Contacts between "dark" cortical epithelial cells and their neighbouring lymphocyte (EC and LC) in 4 months old mice thymus

It is to be noted that not all "dark" and "light" thymus epithelial cells have close spatial relations with their neighbouring lymphocytes on the corresponding ultrathin cuts. Such close relations are most of all characteristic of the cortical epithelial cells and are quite rare as far as medullar cells are concerned. Despite of the special attention paid no morphological data of close relations between the cortico-medullar epithelial cells and polarized medullar epithelial cells with their neighbouring thymocytes are found out.

The electron microscope observations of mice thymus sections rather often show various relations of spatial proximity between epithelial cells and lymphocytes. The aforementioned relations type 1 and 2 are established by other authors, too, in thymus of mice of other breeds, rats and guinea pigs (9, 14, 15, 18, 21) and are usually interpreted as a morphological basis for functional relationship between the epithelial cells and the thymocytes (9, 21). Beside these 2 types in a conventional mice thymus we discerned 2 more types of morphological relations between these cells designed as type 3 and 4, respectively which are not discussed by other investigators.

The data obtained give us reason to suppose that there is a direct functional relationship between cortical epithelial cells and thymus lymphocytes (it is favoured by the general construction of the epithelial cell network in this region). Our data are also in accordance with the peculiarities of the TNC phenomenon

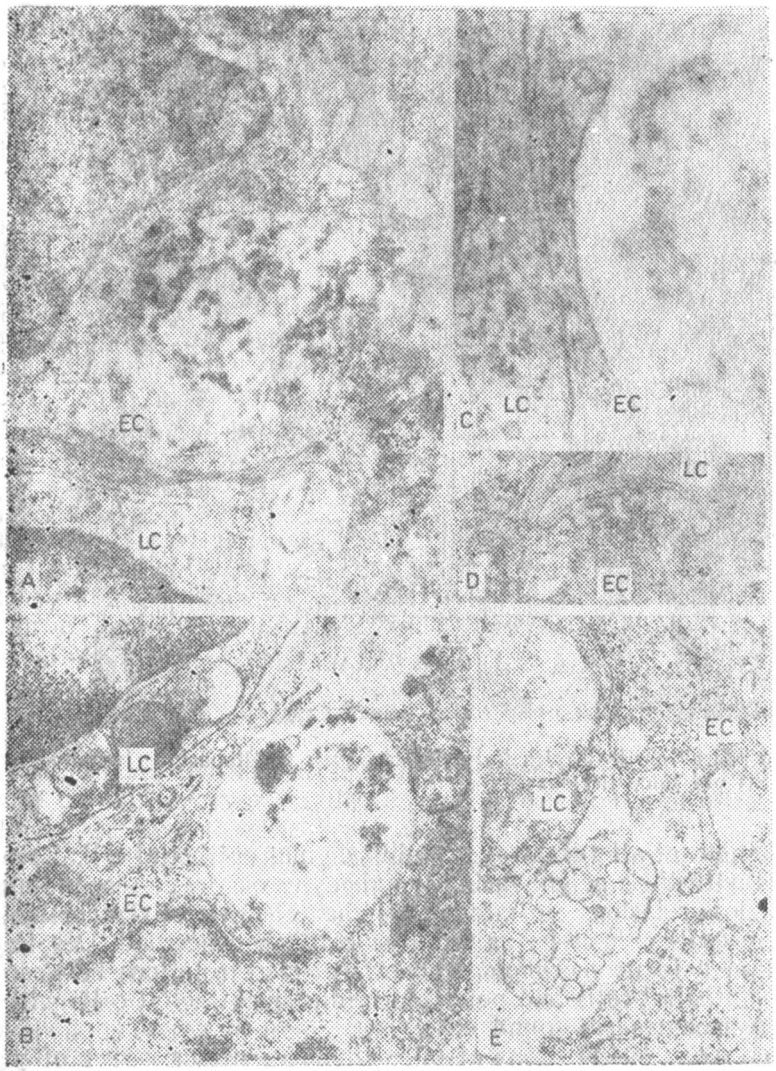

Fig. 2. Contacts between "light" cortical epithelial cells (EC) and their neighbouring lymphocytes (LC) in 4 months old mice thymus

observed in cortical epithelial cells of the thymus of different rodents and men. It is supposed that this phenomenon is crucial for the process of intrathymic T-lymphocyte differentiation (23, 24). Probably, the structural features of the cortico-medullar zone and the medulla of the thymus themselves determine the

possibility of influencing the lymph cells more through a local humoral effect, than through a direct cellular contact.

There also exist differences in the morphology of the epithelio-lymphocyte relationship within the same zone. The "dark" secreting epithelial cells described in the cortex and the medulla possess synthetizing structures for exportation. It seems that in these cells the content of RER, which is most probably of protein nature, is transported to the cellular membrane and is exocytosed within the extracellular matrix by means of channels and vesicles of the RER. The mechanisms of synthesis and exportation of substances from this type of epithelial cells is very similar to the well-known mechanism of protein synthesis and secretion effectuated without the involvement of the Golgi-complex (8), the latter is observed when THF from the antigen-receptors and — transmitting cells in the thymus of guinea pig (22) and in some endocrine cells of the pancreas (5) and the adrenal glands (1) is released.

Probably, the protein producing "light" epithelial cells successfully substantiate the initial steps of the process of protein synthesis and secretion (16). The condensing vacuoles of these cells seem to be the place of initial stocking of the proteins synthesized within these cells. In most epithelial cells a following formation of secretion granules is not observed. But by the condensing vacuoles approaching and sticking to the cell membrane the possibility arises of the vacuolar content being released. We suggest that this process is made easier by the microfibrils, smooth and coated vesicles located in the proximity of the condensing vacuoles and is analogous to the mechanism of intracellular transport of secretory proteins discussed by A. Tartakoff and P. Vossalli (1978). Our description of the possibilities for releasing the epithelial cell synthesized substances— through diffusion and through opening of condensing vacuoles into the intercellular space — is rather similar to the well-known mechanisms of releasing of pancreatic (5) and gastrointestinal (4) hormones, of thyroglobulin (7) and parathyroid hormone (1). At the same time our results agree with the experimental investigations about the synthesis in thymus epithelial cells of THF of different biological action some of which are characterized as hormones (2, 7, 10), as well as with the basically established polypeptide-protein nature of THF (6, 10).

We interpret the data shown for the relationships between epithelial and lymph cells as different mechanisms of synthesis of protein substances, taking place at different stages during the realization of this synthesis. On the grounds of our data we consider that epithelial cells influence lymphocytes inside the thymus directly as well as humorally — by producing locally acting factors.

The features of the morphology of epithelio-lymphocyte relationships described above can be observed in the investigated animals of all ages, but their structural variations are most strongly expressed in the 3—4 months old animals. This is most probably related to the pronounced morphological heterogeneity of thymus epithelial cells in the animals of this age group.

REFERENCES

1. Гордиенко, В. М., В. Г. Козырицкий. Ультраструктура желез эндокринной системы. Киев, Здоров'я 1978, 287 с. 2. Кемилева, З. Тимус. С., Медицина и физкултура, 1979, 231 с. 3. Маринова, Ц. Канд. дис. Варна, 1982. 4. Папазова, М., И. Алтъпармаков. В: Актуални проблеми на медико-биологичните науки. С., 1976, 200—227. 5. Петков, П. Структура и функция на ендокринния панкреас. С., Медицина и физкултура, 1976, 177 с. 6. Петров, Р. В. Имунология и имуногенети-

ка. С., Медицина и физкультура, 1978, 298 с. 7. Розен, В. Б. Основы эндокринологии. М., Высшая школа, 1980, 343 с. 8. Уэйли, У. Аппарат Гольджи. М., Мир, 1978, 244 с. 9. Bartel, H., L. Cieciura. *Acta Histochem.*, 17, 1976, 53—55. 10. Comsa, J. *Med. Welt*, 31, 1980, N 15, 533—536. 11. Jordan, R. K. *J. Anat.*, 132, 1981, N 3, 441. 12. Kendall, M. D. *J. Anat.*, 132, 1981, N 3, 440. 13. Marinova, Tz., E. Boshnakova. *Scr. Sci. Med.*, 16, 1979, 37—42. 14. Mandell, T., *Z. Zellforsch.*, 92, 1968, 159—168. 15. Murray, R. G., et al. *Anat. Rec.*, 151. 1965, N 1, 17—40. 16. Palade, G. *Science*, 189, 1975, 347—358. 17. Reynolds, E. S. *J. Cell. Biol.*, 17, 1963, 208. 18. Scheiff, J. M. *Cell. Tissue Res.*, 170, 1976, N 3, 305—314. 19. Singh, J. *J. Anat.*, 132, 1981, N 3, 444. 20. Tartakoff, A., P. Vossalli. *J. Cell. Biol.*, 79, 1978, N 3, 694. 21. Törö, J., J. Oláh. *Acta morphol. Acad. Sci. Hung.*, 14, 1966, 275—290. 22. Törö, J., J. Oláh, Z. Kiffner. *Acta morphol. Acad. Sci. Hung.*, 28, 1980, N 4, 357—391. 23. Wekerle, H., U. P. Ketelsen. *Nature*, 283, 1980, N 3745, 402—404. 24. Wekerle, H., U. P. Ketelsen. *J. Experim. Med.*, 151, 1980, N 4, 925—944.

МОРФОЛОГИЧЕСКИЕ ОСОБЕННОСТИ ВЗАИМООТНОШЕНИЙ МЕЖДУ ЭПИТЕЛЬНЫМИ КЛЕТКАМИ И ЛИМФОЦИТАМИ ЗОБНОЙ ЖЕЛЕЗЫ МЫШЕЙ

Ц. Маринова, Э. Бошнакова

РЕЗЮМЕ

В результате исследования электронномикроскопическим методом сделана морфологическая характеристика эпительно-лимфоцитарных взаимоотношений в зобной железе конвенциональных мышей различного возраста (новорожденных, инфантильных, половозрелых). Описано четыре типа контактов между эпительными клетками и лимфацитами зобной железы. Отмечаются региональные и возрастные различия в их ультраструктурной характеристике. Проведена интерпретация полученных данных в связи с участием эпительных клеток в процессе внутритимусного Т-лимфоцитарного дифференцирования.