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Development of Lymphocyte Populations in Lymph Nodes of the Mouse

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Lymphocytic colonization of lymph nodes in mouse is studied. Lymphocytes are first seen through the walls of post-capillary venules in diapedesis and nodal parenchyma on the second day after birth. Large lymphocytes are observed in nodal parenchyma between the 4th and the 6th day. The nodular pattern appears on the 24th day with development of primary nodules.

The existence of central and peripheral lymphatic organs is generally accepted. Central organs process stem lymphatic cells, which migrate as T or B lymphocytes to peripheral organs, where lymphocyte populations are built. The moment of arrival of lymphocytes to the peripheral organs and the manner of development of lymphatic populations thereafter were worth investigating.

The present paper studies the colonization of lymph nodes in mouse during the first few days after birth.

Materials and Methods

Forty inbred mice were killed by decapitation at different periods after birth. The fat tissue of axillary and inguinal spaces was dissected. Since lymph nodes are so small at birth that it is impossible to identify them macroscopically, organs from the thoracic cavity were also taken.

The whole thoracic organs were embedded in paraffin for light microscopy and cut in serial sections.

For electron microscopy axillary, inguinal and mediastinal fat tissue was embedded in Epon 812. Semithin sections of 1 μm thick were made and stained with methylene blue and Giemsa and studied with light microscope. When a lymph node appeared, sections about 200 Å were made for the electron microscope.

Results

The early fetal lymph nodes develop by invagination of reticular tissue into the lumen of a lymph channel, which becomes the subcapsular sinus (fig. 1).

At birth lymph nodes are made of high cellular reticular tissue with many blood vessels and numerous nerve bundles. Postcapillary venules are made of basophilic endothelium with or without lumina (figs. 2, 3). The electron microscope shows an incomplete basement membrane surrounding the endothelium. Mitotic figures were often seen in the endothelial cells, as well as some pericytes. The silver stain shows a scarce argyronphylic reticulum.

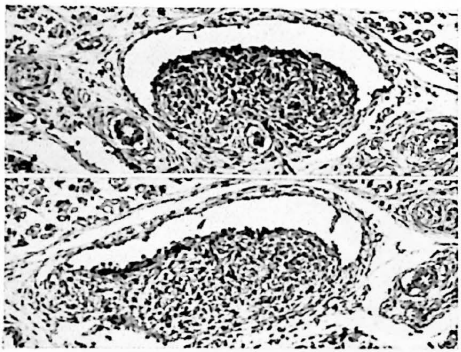

Fig. 1. *Developing lymph nodes of 15 day old fetuses.*
A reticular tissue invaginates in a lymph channel (H. & E. × 90).

Colonization begins on the second day, when lymphocytes are first seen in lumen and then, by diapedesis, in the walls of postcapillary venules. Lymphocytes are practically the only blood cells in these developing postcapillary venules from the 4th to the 6th day.

The migrating lymphocytes are lodged in the extracellular space, but bulging into the cytoplasm of an endothelial cell they appear to be intracellular (fig. 4). On the second day, few small lymphocytes are seen in the nodal parenchyma (fig. 5) whereas on the 6th day lymphocytes are more numerous there than in vessels (fig. 6). There are no nodular patterns as yet. Lymphocytes are spread in the parenchyma singly or in small groups.

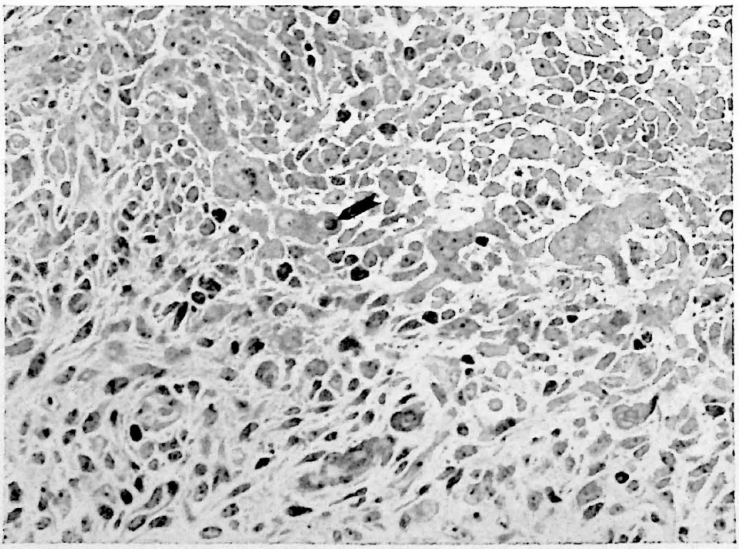

Fig. 2. *Lymph node of a 2 day old mouse.*
Loose reticular tissue, rich in postcapillary venules in development; one contains a lymphocyte (arrow). Semithin section. × 330.

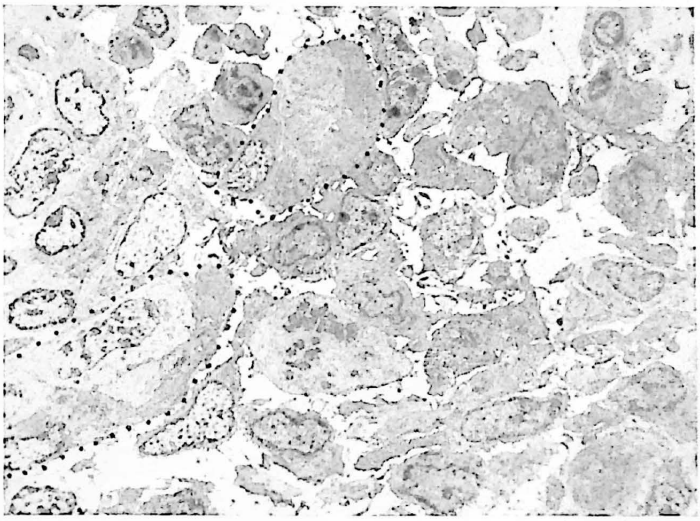

Fig. 3. *Panoramic electron micrograph of a lymph node of 1 day old animal.* There are two vascular bunds without lumen (× 1,300).

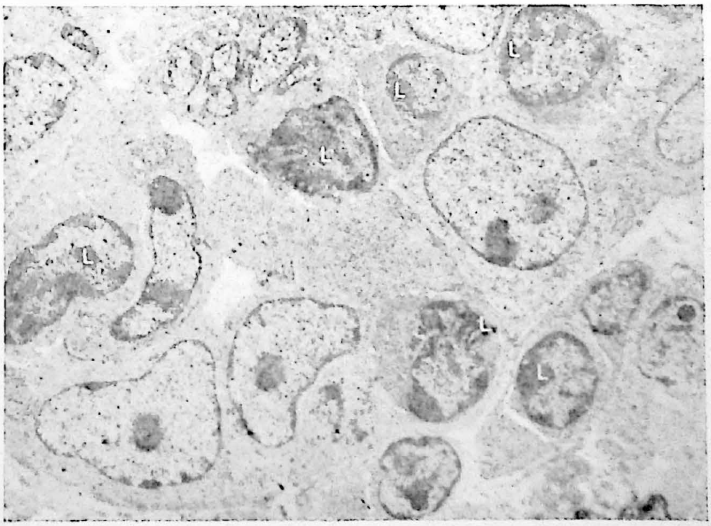

Fig. 4. *Postcapillary venule containing lymphocytes (L) in lumina and in diapedesis (× 3,000)*

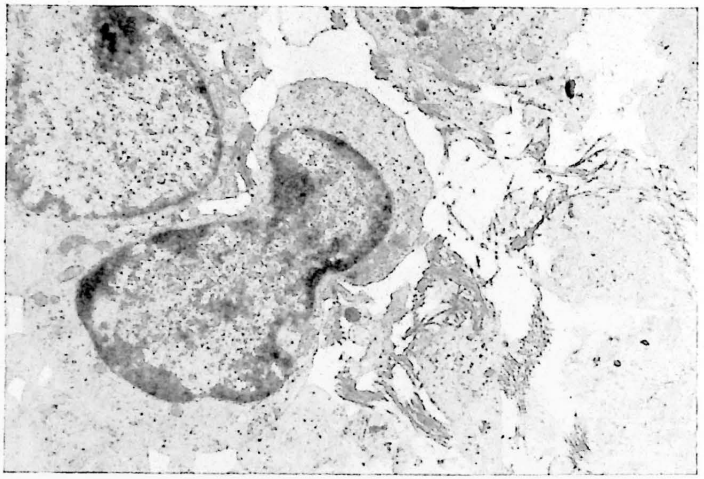

Fig. 5. *Lymphocyte free in the lymp node parenchyma of a 2 day old mouse (× 9,150).*

Lymphocytes are recognized by their size, electron density and lack of organelles, notwithstanding an occasional Golgi Complex and a few mitochondria. The nucleus may be very irregular with many cytoplasmic infoldings (fig. 4). The electrodensity, caused by an abundance of ribosomes, free in cytoplasm or associated to the nuclear membrane, helps to distinguish lymphocytes from endothelial cells. The nucleus of the latter is surrounded by a thin layer of cytoplasm and bulges into the capillary lumen in this period of development. Endothelial cells are further distinguished by their rough endoplasmic reticulum and the frequent presence of desmosomes.

From the 4th to the 6th day large

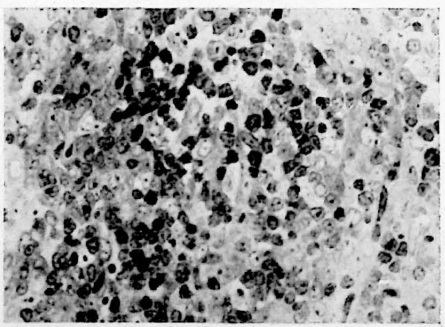

Fig. 6. *Lymph of a 6 day old mouse.* Numerous lymphocytes in parenchyma (×370).

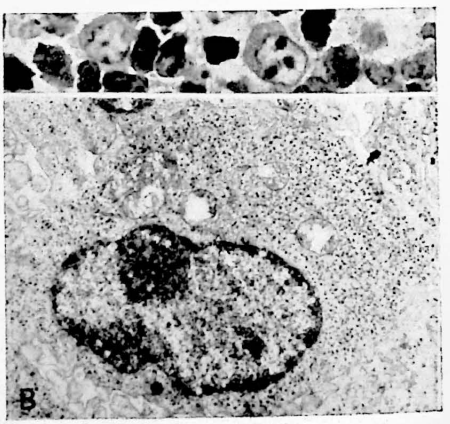

Fig. 7. *Large lymphatic cells in nodal parenchyma of a 4 day old mouse.* Semi-thin section (Giemsa × 650). *B:* Electron mycrograph (× 7,800).

cells with abundant pyroninophilic cytoplasm and prominent nucleolus develop (fig. 7 A). On the electron microscope they appear with a ribosome rich cytoplasm, a few mitochondria with a very clear matrix, and a nucleus with loose chromatin and a prominent nucleolus (fig. 7 B).

In 12 day old animals the cortex and the medulla in lymph nodes are well delimited (fig. 8). The cortex contains many lymphocytes by then, but there is no nodular pattern until the 24th day, when primray nodules without germinal centers develop (fig. 9).

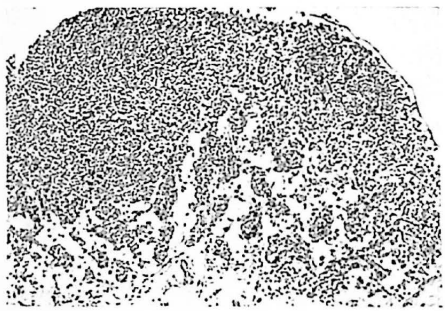

Fig. 8. *Lymph node of a 12 day old mouse.* Well developed medulla and cortex (H. & E. × 75).

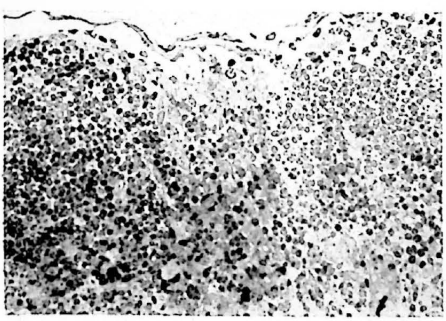

Fig. 9. *Densely packed lymphocytes forming two primary nodules.* 24 day old mouse. Semi-thin section (Giemsa, × 200).

Discussion

These results lead to the conclusion that small lymphocytes begin to arrive at the lymph nodes about the second or third day after birth. They migrate to the nodal parenchyma, where they probably increase in number not only from new arrivals, but also from the *in situ* proliferation of the existent ones. The medium-sized lymphocytes become large pyroninophilic cells and undergo mitosis. Lymphocytes arrive at lymp nodes along with the blood stream and penetrate the wall of postcapillary venules to reach the extravascular space at the 2nd day after birth. SÖDERSTRÖM (8) points out that postcapillary venules are vey important in colonization, but considers that their development in mice takes place on the 9th day after birth. MILLER (7) observes postcapillary venules on the 6th day of life on paraffin sections. In semi-thin sections of material embedded in Epon it is possible to observe postcapillary venules in lymph nodes of 1 day old mice (5).

The lymphocyte colonization of the lymph nodes was studied by MILLER (7) who injected intravenously tritiated thymide labeled lymphocytes. He observed the migration of labeled lymphocytes into the lymph node parenchyma, where the number of lymphocytes increased between the 2nd and the 6th day.

In cat, man and rabbit the arrival of lymphatic cells to lymp nodes occurs before birth (1-4, 6).

On morphological basis it is impossible to establish where the arriving lymphocytes originate, whether from the thymus or elsewhere.

Resumen

Se ha estudiado el proceso de colonización de los ganglios linfáticos por linfocitos, hecho que se inicia el segundo día de vida, cuando los linfocitos se ven atravesando las paredes de las vénulas postcapilares y en el parénquima del ganglio. Entre los días 4 y 6 de vida se

observan ya linfocitos grandes en el ganglio. El patrón nodular aparece hacia el día 24, cuando se forman los folículos primarios (carentes de centros germinativos).

References

1. ACKERMAN, G. A.: *Anat. Rec.*, **158**, 387-399, 1967.
2. BAILEY, R. P. and WEISS, L.: *Anat. Rec.*, **178**, 302, 1974.
3. BAILEY, R. P. and WEISS, L.: *Am. J. Anat.*, **142**, 15-28, 1975.
4. BAILEY, R. P. and WEISS, L.: *Am. J. Anat.*, **143**, 43-58, 1975.
5. DEURS, B. and RÖPKE, C.: *Anat. Rec.*, **181**, 659-678, 1975.
6. HOSTETLER, J. A. and ACKERMAN, G. A.: *Am. J. Anat.*, **241**, 57-76, 1969.
7. MILLER, J. J.: *Lab. Invest.*, **21**, 484-490, 1969.
8. SÖDERSTRÖM, N.: *Scand. J. Hemat.*, **4**, 411-429, 1967.