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Lymphoid Tissue in the Pineal Region of the Mouse: A Histological and Histometrical Study*

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Summary. In 229 dd-mice of both sexes ranging in age from birth to 300 days, the occurrence of lymphoid tissue in the pineal region was examined histologically and histometrically.

In adult mice, the lymphoid tissue appeared usually as small nodular accumulations of small lymphocytes posterior to the pineal stalk. It was commonly localized in close relation to the venous vessels which, arising from a vein from the midbrain, run upwards along the posterior border of the pineal. In addition to lymphocytes, the lymphoid tissue contained varying numbers of plasma cells which tended to increase in number with age.

The lymphoid tissue was not found in mice younger than 10 days. At 15 days it first occurred as a small patch of small lymphocytes in 11% of the cases examined. Thereafter, the lymphoid tissue increased in size and amount rapidly with advancing age. After 60 days the lymphoid tissue commonly appeared as a nodular accumulation of lymphocytes. In each age group after 60 days the lymphoid tissue was found in more than 90% of the cases examined. The nodular lymphoid tissues measuring 50 to 150 μ in diameter were most frequent, but the larger ones tended to increase in number as age increased. The lymphoid tissue tended to increase in total amount with advancing age.

As is generally known, no lymph nodes are present in the central nervous system and its meninges (HAM, 1969). However, little information is available in the literature as to whether there may occur lymphoid tissue in the central nervous system in mammals. In birds, lymph nodules have been microscopically found near and in the pineal (ROMIEU and JULLIEN, 1942; SPIROFF, 1958; QUAY, 1965).

During the course of studying the postnatal development of the mouse pineal, we observed the frequent occurrence of lymphoid tissue in the pineal region. Since this tissue was of particular interest because of its intracranial position, we attempted systematically to examine its development during life, qualitatively and quantitatively.

Material and Methods

In this study, a total of 229 dd-mice of both sexes were used, which were maintained under constant environmental conditions and given *ad libitum* commercial food pellets (NMF, Oriental Enz. Co., Japan) and water. They were killed at ages of 0, 10, 15, 20, 30, 40, 60, 90, 120, 200 and 300 days. In each age group 15 to 30 animals were used from each sex. At the scheduled age, the mice were killed with chloroform and the pineal was immediately removed with the surrounding brain tissues, to be fixed in Bouin's fluid for 24 hours. After being embedded in paraffin, the tissue was serially sectioned at 6 μ in the sagittal plane of the pineal. The sections were stained

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usually with hematoxylin and eosin and often with periodic acid-Schiff reagent (PAS) and hematoxylin.

Results

1. Distribution of veins in the pineal region

In the pineal region, lymphoid tissue, when present, was found in close topographical relation to veins. The venous distribution in the pineal region of the mouse, which is diagrammatically shown in Figure 1, was made clear on the basis of reconstructive examination from serial sections in adult mice. Its basic pattern was similar to that of the rat (KAPPERS, 1965), although the course and development of each vein showed some variation among individuals. Immediately above the distal tip of the pineal body a superior sagittal vein joins two paired transverse sinuses at the confluens sinuum. Several other veins also converge into the confluence. Two great cerebral veins (vena cerebri magna) run upwards closely along the lateral aspects of the pineal and empty into the confluence. Each of the paired great cerebral veins receives two major veins; a basal vein arising from the thalamic region and an internal cerebral vein receiving choroidal veins from the choroid plexuses in the lateral ventricles. Either the right or left side of the basal veins also receives a small vein draining the midbrain, and the internal cerebral vein receives an internal sagittal vein from forwards. The paired basal veins communicate with each other at the level of the pineal stalk. Posteriorly along the pineal body an anastomotic vessel passes upwards from the vein from the midbrain to the confluens sinuum.

Fig. 1. Diagram of venous distribution in the pineal region. *P* pineal; *BV* basal vein; *CS* confluens sinuum; *CV* choroidal vein; *GCV* great cerebral vein; *ICV* internal cerebral vein; *ISV* Internal sagittal vein; *OV* occipital vein; *SSV* superior sagittal vein, *TS* transverse sinus.

Into the sinus confluence an occipital vein from the cerebellum also empties from backwards.

Fig. 2. A nodular accumulation of lymphocytes. The accumulation is located close to the venous wall posterior to the pineal stalk. 120-day-old male; Bouin-fixation; PAS and hematoxylin $\times 140$.

Fig. 3. Diagram of the position of lymphoid tissue at various age. Black dots represent nodular accumulations of lymphocytes.

2. Position of lymphoid tissue in the pineal region

Lymphoid tissue in the pineal region was generally located just behind the pineal stalk and close to the confluence of a few veins arising from the midbrain (Fig. 2). In addition, lymphoid tissues occurred also in the other positions, such as just above the pineal body, anterior to the stalk and anterior to the internal cerebral veins, particularly in relatively aged mice (Fig. 3).

3. Structure and development of the lymphoid tissue

In adult mice, the lymphoid tissue in the pineal region appeared usually as spherical, dense collections of small lymphocytes (Fig. 2, 4). There were also small numbers of larger lymphocytes dispersed among small lymphocytes. Mitotic figures, though rarely, were seen among larger lymphocytes (Fig. 4). There were also occasional macrophages, which sometimes contained cell debris within the cytoplasm. In the periphery of the lymphoid tissue there were often plasma cells in various stages of maturation (Fig. 5).

Fig. 4. Lymphoid tissue in the pineal region. The lymphoid tissue occurs as a dense collection of small lymphocytes. A mitotic figure of larger lymphocytes is seen in the upper part of the figure. 60-day-old male; Bouin-fixation; PAS and hematoxylin. $\times 1,300$

Fig. 5. Plasma cells in the periphery of the lymphoid tissue. 120-day-old female; Bouin-fixation; PAS and hematoxylin. $\times 1,300$

As mentioned above, the lymphoid tissue was usually found close to the venous vessels. The veins were frequently congested with blood cells, in which small lymphocytes were found scattered singly or in small groups particularly toward the adjacent lymphoid tissue.

No lymphoid tissue was visible from birth to 10 days of life, although a few small lymphocytes were scattered around the pineal. Almost all the cases at 15 days of

age possessed scattered lymphocytes near the pineal. At 20 days, small lymphocytes were accumulated in small groups close to the confluence of the venous vessels mentioned above. At first the lymphoid tissue was less dense in the population of lymphocytes, but it gradually became packed densely with lymphocytes as the age advanced. The size and number of the accumulations of lymphocytes also increased gradually with progressing age. After 60 days the lymphoid tissue always occurred as a nodular accumulation of small lymphocytes. In addition, diffuse lymphocyte infiltration close to the venous wall showed a tendency to increase. Plasma cells also increased in number with age.

4. Quantitative findings of the growth of the lymphoid tissue

As mentioned above, no lymphoid tissue appeared until 10 days of age. At 15 days a small patch of lymphocytes first occurred in 2 out of 19 cases examined.

Table 1. Frequency of the occurrence of cases with lymphoid tissues for each age period.

Days of age	0	10	15	20	30	40	60	90	120	200	300
Number of mice	15	27	19	16	27	21	20	12	21	30	21
Number of cases with lymphoid tissue (%)	0 (0%)	0 (0%)	2 (11%)	9 (56%)	17 (63%)	15 (71%)	18 (90%)	11 (92%)	19 (90%)	28 (98%)	21 (100%)

Thereafter, the lymphoid tissue developed rapidly with age. The frequency of the occurrence of the lymphoid tissue for each age period is presented in Table 1. As seen in this table, occurrence rapidly increased after 20 days. The lymphoid tissue was found in more than 90 % of the cases for each age period after 60 days. At 300 days all the cases examined possessed lymphoid tissue in the pineal region. The nodular lymphoid tissues measuring 50 to 150 μ in diameter were most common, but the larger ones tended to increase in number as age progressed (Fig. 6). In this figure, the size of each nodule expressed as the largest sectional area which was measured by the longest and shortest diameters of the nodule. As seen in Figure 7, larger nodules, measuring larger than 150 μ in diameter, were found in under 15 % of the lymphoid tissues examined up to 60 days, but then they increased with age until at 200 and 300 days they occupied about 40 %.

The number of the nodular lymphoid tissues was counted in each case for every age. Its frequency distribution expressed as a percentage is presented for each age (Fig. 8). As seen in this figure, a nodule, if present, is single in mice younger than 15 to 20 days old. At 30 days there were a few cases with two nodules. After 60 days the cases with two nodules or more increased in number.

The total size of the lymphoid tissues was calculated by both the number and the area for each case in each age group (Fig. 9). This figure indicates that the lymphoid tissues also grew in total amount with advancing age.

Discussion

In the body there are generally found small collections of lymphocytes as well as lymph nodules in many places (HORII and KAWAKAMI, 1963). Many such small lymphoid tissues are thought to be transitory structures which may come and go

Fig 6. Size of lymphoid tissue at various ages. Bars represent means.

Fig. 7. Frequency distribution of the occurrence of lymphoid tissues of less than 150 μ and more than 150 μ in diameter at various ages.

(ANDREW, 1971). As already mentioned, however, little information is available as to the presence of intracranial lymphoid tissue in mammals. In birds, a lymph nodule is often found in and near the pineal (ROMIEU and JULLIEN, 1942; SPIROFF, 1958; QUAY, 1965). In the domestic fowl, ROMIEU and JULLIEN (1942) discovered patches of lymphocytes between the posterior part of the pineal and the meninges and further also within the gland, and they considered the lymphoid tissue as the source of lymphocytes for the meninges and cerebrospinal fluid. Subsequently, SPIROFF (1958) also observed that in the post-hatching stages, beginning 12 days after hatching, lymphocytes invade the pineal progressively and form lymph nodules on the walls of major blood vessels within the pineal. The size of these lymphoid areas within the pineal reaches a maximum at three months after hatching, and then gradually decreases and finally disappears in the adult pineal. According to QUAY (1965) there are nearly always pineal lymph nodules within either the capsule or the internal connective tissue stroma even in one-year-old fowls.

The observations described in this report confirm the presence of lymphoid tissue in the pineal region of the mouse. The lymphoid tissue is most commonly located in close relation to certain veins posterior to the pineal stalk, and it is found in more than 90 % of the cases examined in both sexes after 60 days of age. From the present findings of its development during life it is most likely that the lymphoid tissue occurs physiologically and consistently in this region.

Fig. 8. Frequency distribution of the numbers of lymphoid tissues at various ages. Open bar, without lymphoid tissue; hatched bar, with lymphoid tissue.

Fig. 9. Total size of lymphoid tissue at various ages. Bars represent standard deviations.

At present, the brain as well as the anterior chamber of the eye and the testis is considered to be an efficient site for tissue implantation (GREENE, 1967; WALTER and ISRAEL; 1970), because the tissues transplanted survive relatively longer in these sites than elsewhere. As for the brain it is generally thought that since it has no lymphoid tissue and possesses the blood-brain barrier, intracranial grafts elicit slow immune responses. On the other hand, however, it is said that the intracranial site is not always efficient for tissue implantation (TANAKA, ORITA and KOKUMAI, 1970). In this relation, the almost constant occurrence of lymphoid tissue in the pineal region is of interest, because the relative efficiency, if present, of the brain for tissue implantation may not be explained only by the absence of the intracranial lymphoid tissue at least in the mouse. In view of the recently accumulated knowledge about the functional significance of lymphoid tissue (for review see YOFFEY and COURTICE, 1970), the presence of lymphoid tissue in the pineal region may be associated, in some way, with immune responses in the intracranial site, but its exact significance remains uncertain at present.

As is the case of the fowl, the lymphoid tissue in the pineal region of the mouse is also characteristically present and its distribution and development are age-dependent. In agreement with the view of QUAY (1965), however, there is no definite evidence to support the concept of ROMIEU and JULLIEN (1942) that the lymphoid tissue serves as the source of lymphocytes of the meninges and the cerebrospinal fluid, although the possibility can not absolutely be excluded.

マウス松果体の周辺におけるリンパ組織—組織学的ならびに計量組織学的研究

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新生仔期から生後300日齢までの雌雄229例の dd-マウスを用いて、頭蓋内 とくに松果体の周辺におけるリンパ組織の分布と発育を、組織学的ならびに計量組織学的に検索した。

リンパ組織は主として密在する小リンパ球の小節状集積からなる。リンパ組織には形質細胞もしばしばみとめられ、かつ加齢とともに増加する傾向がある。リンパ組織の出現部位は松果体周辺の静脈と密接な関連がみられ、とくに松果体茎の後側で中脳からの静脈が合流する部位に最も多い。

リンパ組織は生後10日までは見られないが、15日で11%の例に小集積として始めて出現し、その後急速に発達し、60日以後では90%以上の例に出現する。

リンパ組織の小節状集積は一般に径 50~150 μ を算するものが最も多いが、さらに大きなものも加齢とともに増加する。またその総量も加齢とともに増加する。

References

- Andrew, W.: The anatomy of aging in man and animals. New York-London, Grune and Stratton, 1971.
- Greene, H. S. N.: The use of transplanted tissues in biology and histology. In: (ed. by) G. Bourne: In vitro techniques in histology. Baltimore, Williams and Wilkins, 1967. (p. 80-112).

- Ham, A. W.:** Histology. 6th ed. Philadelphia-Toronto, T. B. Lippincott Co., 1969. (p. 518).
- Horii, I. and Y. Kawakami:** Distribution and development of lymph nodes (Japanese). In: (ed. by) Japan Hematological Society: Nippon Ketsuekigaku Zensho, Vol. 1. Tokyo, Maruzen, 1963. (p. 797-822).
- Kappers, J. A.:** Survey of the innervation of the epiphysis cerebri and the accessory pineal organs of vertebrates. Progr. Brain Res. 10: 87-151 (1965).
- Quay, W. B.:** Histological structure and cytology of the pineal organ in birds and mammals. Progr. Brain Res. 10: 49-86 (1965).
- Romieu, M. et G. Jullien:** Sur l'existence d'une formation lymphoïde dans l'épiphyse de Gallinacés. C. r. Soc. Biol. 136: 626-628 (1942).
- Spiroff, B. E. N.:** Embryonic and post-hatching development of the pineal body of the domestic fowl. Amer. J. Anat. 103: 375-481 (1958).
- Tanaka, S., K. Orita and Y. Kokumai:** Transplantation immunology (Japanese). Tokyo, Asakura-Shoten, 1970.
- Walter, J. B. and M. S. Israel:** General pathology. 3rd ed. London, J. and A. Churchill, 1970. (p. 312).
- Yoffey, J. M. and F. C. Courtice:** Lymphatics, lymph and the lymphomyeloid complex. London-New York, Academic Press, 1970.

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