On the Lymphatic Vessels in the Stroma of the Cornea

By

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Using several textbooks, the authors searched for the description on the lymphatic vessels in the stroma of the cornea. But those books of Rauber-Kopsch (1955), Axenfeld (1958), Stöhr et al. (1959), Tokari (1960), Hagiwara et al. (1961), Nishimura et al. (1962), Suda et al. (1962), Adler (1963), Bailey (1964), Kaneko (1964), Möllendorff (1964), Okajima (1964), Mori-Ogawa (1966), Gray (1967), Bloom-Fawcett (1968), Ham (1969), Mori et al. (1969), and Omochi et al. (1970) described nothing or did not affirm the existence of those vessels. Von Recklinghausen (1888) investigated on the canaliculi in the cornea of the dead but he did not find out the lymphatic vessels in that organ. However, regarding this report, Gerlach (1888) discussed that he could find the canaliculi which were lined with endothelial cells in the cornea of the swine and the newborn baby after the treatment of the silver impregnation. But he did not call those canaliculi as the lymphatic vessels. Hayakawa (1943) made experimental keratitis with injection and inoculation of tuberculous bacilli, staphylococcus, and vaccine. He has minutely investigated on the changes of the corneal spaces and their processes in those objects, but his report did not say anything about the canals in the stroma. After injecting trypan blue and victoria blue 4R into the stroma, Ishii (1954) observed the fluid flowing in the stroma of the rabbit. Moreover, she has cauterized the cornes of the frogs with nitrate of silver and then stained those materials with silver and toluidine blue. But for those experiments, she did not touch upon the canal system in the stroma of rabbits and frogs. Aoki (1949) denied the existence of the true lymphatic vessels in the normal cornea but through his experiments, using silver nitrate on the cornes of rabbits, blood capillaries and true lymphatic vessels were newly formed in the cornea. Katayama (1951) also recognized newly formed blood vessels and lymphatic capillaries in the cornea of rabbits and man by his experiments. Afterwards (1957) he injected a small quantity of
0.3% H₂O₂ solution into the corneas of removed ox eyeballs and found canaliculi in the stroma which were filled with gas. He believed those canaliculi were expanded lymphatic vessels. But he could not recognize the endothelial cells at the walls of the expanded spaces with the histological research, while the lymphatic vessels in the conjunctiva had the endothelial cells. Moreover, he practiced the same experiments with cocainized corneas of living rabbits and man and obtained the same view as with the ox.

As the present authors have confirmed the true lymphatic vessels in the stroma of the cornea of many kinds of animals, they will inform on the result in this report.

Materials and Methods

Ten bullfrogs, twelve rats, one cat, four rabbits, six dogs, three swines, and one Japanese monkey were used for the present study. The removed eyeballs were soaked after removal of the epithelial cells of the cornea in 0.25% AgNO₃ solution to practice the silver impregnation of the corneas. The times for the impregnation were as follows. Bullfrog 2–3 minutes, rat 3–5 minutes, cat and rabbit 8–10 minutes, dog and swine 10–13 minutes, monkey 17–20 minutes. After that the eyeballs were washed for several minutes with distilled water and were fixed for about 12 hours with buffered formaline. And then the corneas were cut off from the eyeballs and stained for 3–7 days with alaun carmine. Namely, the bullfrog and the rat for 3 days and the rest of the animals for 5–7 days. After staining the corneas were passed through alcohols of increasing to absolute, and then into xylol. Last of all, they were mounted in xylol-balsam. At that time, the corneas of big animals were cut into several pieces.

Several canaliculi always became visible in the stroma of the cornea after the silver impregnation. Several pieces of corneas of bullfrogs and rabbits, including such canaliculi, were cut off and those pieces were embedded in celloidin and cut in 6μ thin cross sections of the canaliculi.

Observations

With the silver impregnation, not only numerous corneal spaces and their processes but several canaliculi appeared in the stroma of the corneas of all animals which were observed by the authors (Figs. 1, 2). The corneal spaces and their processes open into the canaliculi here and there (Fig. 2), and those canaliculi communicate with the
lymphatic capillary network formed in the stroma near the corneal margin (Fig. 3), and the network is connected with the lymphatic vessels in the scleral conjunctiva near the corneal margin. The capillaries in the stroma have small caliber at the beginning but they gradually increase in diameters as they progress to the end. They do not form the network except near the corneal margin. The walls of those canaliculi consist of an endothelial lining (Figs. 5, 6, 7) and the lining is externally surrounded by the latticed fibers (Figs. 8, 9). The latticed fibers and the borders of the endothelial cells appear with the silver impregnation (Figs. 1, 5, 6, 8, 9).

Discussion

As the authors have said above, many textbooks and several articles are positive in denying the presence of the lymphatic capillaries in the stroma of the cornea in man and many kinds of animals. Gerlach (1888) said that he could find canaliculi lined with endothelial cells in the cornea of the swine and the newborn baby with the silver impregnation. But his description is so short that the authors know nothing beyond that. Aoki (1949) defined the lymphatic vessels in the normal cornea but after using silver nitrate on the cornea of the rabbit, he recognized newly formed blood vessel and lymphatic capillaries in it. Katayama (1951) got the same result with rabbits and man. Afterwards (1957), by the injection of weak H₂O₂ solution into the cornea of living rabbits, man, and removed ox eyeballs, he found many canaliculi in the stroma which were filled up with gas. Although he could not histologically prove the endothelial lining of the canaliculi, he considered them as the lymphatic vessels. But depending on his description and figures, the authors can not agree with his opinion. Namely, the canaliculi which were regarded as the lymphatic vessels by Katayama were nothing but expanded corneal spaces and their processes. The very canaliculi which were found by the authors were true lymphatic vessels. Because, the canaliculi communicate with the lymphatic vessels in the conjunctiva and their walls were formed with the endothelial cells and they were externally surrounded by the latticed fibers. The adjacent corneal spaces communicate with each other by their processes, and those spaces and processes open into the canaliculi here and there. Therefore, passing to the corneal margin, the canaliculi are fed by those spaces and processes. Namely, those canaliculi are the lymphatic vessels which gather the tissue fluid contained in the corneal spaces.
Summary

Several canaliculi are observed in the stroma of the cornea of the bullfrog, rat, rabbit, cat, dog, swine, and Japanese monkey. Those canaliculi have an endothelial lining and they are externally surrounded by the latticed fibers. The borders of the endothelial cells and the latticed fibers appear with silver impregnation. The corneal spaces and their processes open into the canaliculi here and there. The authors consider that those canaliculi are the lymphatic vessels which gather the tissue fluid contained in the corneal spaces in the stroma. The canaliculi which were regarded as the lymphatic vessels by Katayama were nothing but expanded corneal spaces and their processes. Because, those canaliculi had no endothelial lining.

References

Gerlach, L. Cited from von Recklinghausen.
Gray's Anatomy 1967. 1285, 1286.
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PLATES
Explanation of Figures

Plate I

Figs. 1, 2. Lymphatic vessel in the stroma of the cornea of the rabbit. †: corneal spaces and their processes. Silver impregnation. ×240.

Fig. 3. Lymphatic vessel, corneal spaces, and their processes in the stroma of the cornea of the bullfrog. Silver impregnation. ×240.

Fig. 4. Lymphatic capillary network formed in the stroma near the corneal margin of the rabbit. Silver impregnation. ×36.

Fig. 5. Nuclei of the endothelial cells (†) of the lymphatic vessel in the stroma of the cornea of the bullfrog. Stained for 3 days with alaun carmine after silver impregnation. ×400.

Fig. 6. Nuclei of the endothelial cells (†) of the lymphatic vessels in the stroma of the cornea of the rabbit. Stained for 5 days with alaun carmine after silver impregnation. ×400.

Plate II

Figs. 7, 8. Cross section of the lymphatic vessel in the stroma of the rabbit. †: Nuclei of the endothelial cells. A: Silver granules. Stained for 5 days with alaun carmine after silver impregnation. ×720.

Fig. 9. Longitudinal section of the lymphatic vessel in the stroma of the cornea of the rabbit. †: Nuclei of the endothelial cells. Stained for 5 days with alaun carmine after silver impregnation. ×720.

Fig. 10. Longitudinal section of the lymphatic vessel in the stroma of the cornea of the bullfrog. †: Nuclei of the endothelial cells. Stained for 3 days with alaun carmine after silver impregnation. ×720.

Fig. 11. Latticed fibers on the capillary wall (A), and borders of the endothelial cells (B) of the lymphatic vessels in the stroma of the rabbit. Silver impregnation. ×300.

Fig. 12. Latticed fibers on the capillary wall of the lymphatic vessel in the stroma of the cornea of the rat. Silver impregnation. ×400.