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Distribution and Role of Lymph Vessels of the Bursa Fabricii

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Summary

The fine distribution of lymph vessels in the bursa of Fabricius and the efferent lymph vessels coming from the bursa were demonstrated. The lymph vessels never entered lymphoid follicles, whereas the blood vessels pierced the cortex of lymphoid follicles and formed a network at the boundary between the cortex and medulla of lymphoid follicles. The lymph vessels commenced in the immediate exterior of lymphoid follicles. A large number of lymphoid cells were found in the lumen of the lymph vessels. Migration of lymphoid cells from the cortex of lymphoid follicles to the lymph vessels was also observed. Therefore, it was assumed that the lymph vessels of the bursa are important in the transport of lymphoid cells from the bursa to the blood stream.

Introduction

In birds, the bursa of Fabricius has an essential immunological function as a central lymphoid organ (1, 2, 4, 9, 10, 13) by seeding immunocompetent cells to peripheral sites where they become antibody-producing cells or function as a peripheral lymphoid organ (6, 11, 12, 15, 16) for local antibody production. Information about the lymph vessels of the bursa is however completely absent, though the distribution of blood vessels has been demonstrated in detail (14). This paper will present the distribution and role of lymph vessels in the Fabricious bursa.

Material and Methods

Non-inbred White Leghorn chicks, 3-4 weeks of age, were employed for this study. After exposure of the bursa through a midline incision in the abdominal wall in one group of animals, a very small amount of Pelikan ink (C11/1431a, Günther Wagner, Pelikan Werke, Hannover, Germany) was injected by means of a syringe with a fine needle (27G, 0.4 mm external diameter) into the capsule of the bursa at the sites of the immediate vicinity of blood vessels. In the second group, the right and left sciatic arteries and abdominal aorta were ligated, and then about 4-5 ml of Pelikan ink were slowly injected into the median sacral artery at the site of just below the bifurcation of the abdominal aorta to the sciatic arteries (Fig. 1). After injection of Pelikan ink into the median sacral artery, silver nitrate solution, 0.5 percent, was injected in the same manner into the capsule of the bursa at the immediate vicinity of blood vessels containing Pelikan ink. The bursa in both groups was biopsied together with the ureter and the pudendal artery and vein as soon as possible after macroscopic observation and was fixed in 10 percent formalin. The sections at 20 micron thickness were stained lightly with hematoxylin and eosin. In the third group, the non-treated bursa was excised immediately after sacrifice, fixed in Carnoy's fluid, embedded in paraffin wax, sectioned at 8 micron thickness, and then stained with methyl green-pyronin.

Results

Immediately after injection of Pelikan ink into the capsule of the bursa at the sites of immediate vicinity of blood vessels, irregularshaped tubules containing Pelikan ink and forming a plexus by anastomoses appeared on the surface of the bursa. These tubules showed constrictions and dilatations at intervals as a characteristic feature of lymph vessels and were distinctly different from the uniform blood vessels. Blind ends which were also characteristic of the lymph vessels were often observed. Some of lymph vessels ran closely adjacent to the blood vessels and others independently.

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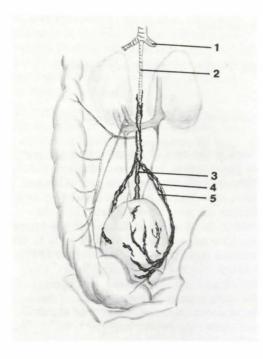


Fig. 1 Scheme of the lymph drainage of the bursa. (1) A. Ischiadica, (2) A. sacralis media, (3) A. caudalis, (4) A. pudenda, (5) V. pudenda

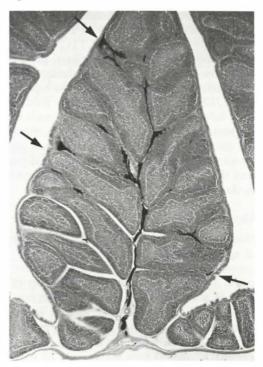
The lymph vessels of the bursa's capsule came together at two places where the right and left pudendal arteries and veins entered the bursa (fig. 1). Two or 3 efferent lymph vessels emerged from the bursa and ran upwards with the pudendal blood vessels to about the level where the median sacral artery branched to the caudal artery and pudendal arteries (fig. 1). They communicated with each other by anastomoses around the pudendal blood vessels and finally entered a part of the sub-

Fig. 2–3 Serial sections of a fold at 20 micron thickness, stained with HE. The lymph vessels containing Pelikan ink distribute in the central connective tissue septum of the fold and in the peripheral connective tissues between lymphoid follicles. The commencement of the lymph vessels in the connective tissues under or near the epithelium is indicated by arrows. The blood vessels contain no ink. x35



Fig. 2

Fig. 3 4



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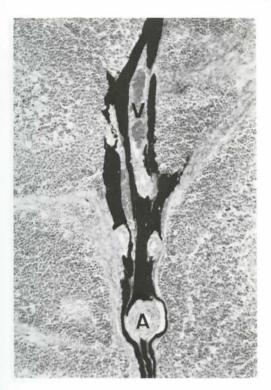


Fig. 4 A section at 20 micron thickness, stained with HE. The tubular lymph vessels containing Pelikan ink are distinguished from the tissue spaces. They form a plexus in the connective tissue septum of the fold. Note the lymph vessels immediately exterior to lymphoid follicles. No ink is found in the lumen of the arteries (A) and veins (V). x70

vertebral trunk (*Hoyer*, 3), running alongside of the caudal aorta or caudal lymph vessels (*Kampmeier*, 5). Besides, Pelikan ink injected directly into the lymph vessels of the bursa's capsule appeared frequently in the lumen of lymph vessels of the cloacal wall next to the bursa. This means that the lymph vessels of the bursa and cloaca communicated with each other.

In the sections of the bursa, irregular, tortuous, tubular canaliculi containing Pelikan ink were found in the connective tissue septa of each grossly visible fold projecting into the bursa's lumen (figs. 2-4). They were clearly distinguished from the following blood vessels

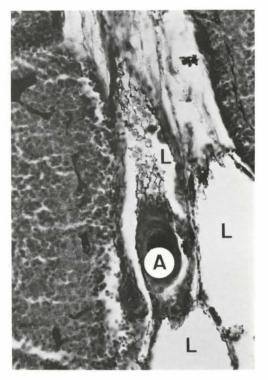


Fig. 5 A section at 20 micron thickness, stained with HE. The intercellular substances of endothelial cells of a lymph vessel (L) surrounding a artery (A) are stained dark brown with silver nitrate. The blood vessels distributing at the boundaries between the cortex and medulla of lymphoid follicles are filled with Pelikan ink. x240

which contained no Pelikan ink. It was thought that these tubles were the lymph vessels into which Pelikan ink entered as a result of backflow. The lymph vessels in the septa of folds formed a plexus by anastomoses around the following blood vessels. The lymph vessels in the septa of folds received a number of tributaries which probably began in the submucous connective tissues and ran towards the septal lymph vessels through the connective tissues between adjacent lymphoid follicles. The staining with silver nitrate solution revealed that the walls of lymph vessels consisted of characteristic endothelial cells (figs. 5, 6). The endothelial cells of the lymph vessels were



Fig. 6 A section at 20 micron thickness, stained with HE. The lymph vessels (L) are observed in the immediately exterior of lymphoid follicles. The endothelial cells of lymph vessels are stained dark brown with silver nitrate. Pelikan ink is injected into the blood vessels. x240

larger than those of the blood vessels and their edges showed a peculiar wavy line.

Lymph vessels were often found in the immediate periphery of lymphoid follicles (figs. 4, 6). However, they never entered lymphoid follicles, though the blood vessels penetrated the cortex of lymphoid follicles and formed a network in the boundaries between the cortex and medulla of lymphoid follicles (fig. 7).

In the thick sections of the bursa injected with Pelikan ink or silver nitrate solution, numbers of cells were frequently observed solely or in a group in the lumen of the lymph vessels. However, it was difficult to identify the type of cells and therefore thin sections stained with methyl green-pyronin were made. The

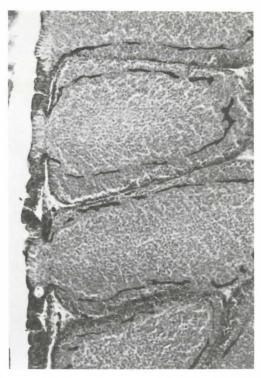


Fig. 7 A section at 20 micron thickness, stained with HE. The blood vessels filled with Pelikan ink form a network in the boundaries between the cortex and medulla of lymphoid follicles. x70

lymph vessels were able to be distinguished from the blood vessels containing nucleated erythrocytes. The cells in the lumen of the lymph vessels were all of lymphoid cell nature in various stages of differentiation similar to those in the cortex of lymphoid follicles (figs. 8, 9). Large numbers of large pyroninophil immature cells were present under normal conditions. Migrating lymphoid cells across the boundary of lymphoid follicles from the cortex to the lymph vessels in the interfollicular connective tissues were also observed (figs. 8, 9).

Discussion

The minute lymph vessels in the parenchyma of the bursa were demonstrated by retrograde

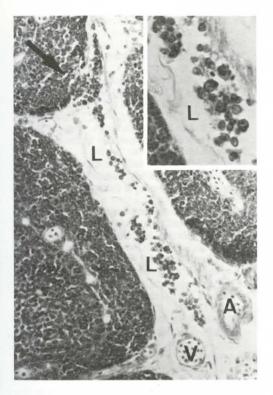




Fig. 8 A section at 20 micron thickness, stained with methyl green-pyronin. A large number of lymphoid cells can be seen in the lumen of a lymph vessels (L) which is distinguished from the artery (A) and vein (V) containing nucleated erythrocytes. Higher magnification (x480) of a part of the lymph vessels at the right upper corner. Note large pyroninophil cells in the lymph vessel. Migration of lymphoid cells from the cortex of a lymphoid follicle is indicated by arrow. x240

Fig. 9 A section at 8 micron thickness, stained with methyl green-pyronin. Lymphoid cells in various stages of differentiation are seen in the lumen of a lymph vessel (L). Migration of lymphoid cells across the boundary of the lymphoid follicle is indicated by arrow. The vein (V) contain many nucleated erythrocytes. x480

injections of Pelikan ink into the lymph vessels of the capsule. Although the capsular lymph vessels were provided with valves, Pelikan ink could reach the parenchymal lymph vessels probably through the by-pass of anastomoses. Injections of some dyes into a lymph vessels of the serosa for filling the lymph vessels of the mucosa can be successful as an orthodox procedure.

The lymph vessels of the bursa never entered lymphoid follicles, though the blood vessels pierced the cortex of lymphoid follicles and formed a network at the boundaries between the cortex and medulla of lymphoid follicles. However, the lymph vessels were located in the immediate periphery of lymphoid follicles. Moreover, large numbers of lymphoid cells were found in the lumen of the lymph vessels under normal conditions. Migration of lymphoid cells from the cortex of lymphoid follicles to the lymph vessels was also observed. Therefore, one of the important roles of the lymph vessels in the bursa appears to be the transport of lymphoid cells from the bursa to the general circulation just as the lymph vessels of the thymus of mammals transport thymocytes from the thymus to the blood stream (7, 8). It should be noted that many large pyroninophilic cells were contained in the lymph vessels of the bursa, although the majority of cells in lymph from the thymus are small lymphocytes. The lymph vessel drainage does not exclude direct transport of lymphoid cells via blood vessels.

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