Comparative histology of human and dog spleen

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Abstract

Introduction: Spleen is the largest secondary lymphatic organ in the body designed to respond immunologically to antigens circulating in blood. Spleen performs important functions such as red blood cells production, their destruction, blood filtration, storage of blood and phagocytosis. Most of the textbooks of microscopic anatomy show picture of human spleen as the combination of findings derived from various animals. **Aim:** The present study was undertaken to compare histological difference between human and dog spleen as they differ according to their functions. **Material and methods:** Five mature dog and five normal human spleens were procured and were kept in Bouin's fluid for fixation. Sections were cut after paraffin embedding. Slides were stained by Haematoxylene and Eosin, Masson's Trichrome, Gordon and Sweet's reticular stain and observed under light microscope. **Results:** Capsule and trabeculae contains collagen and elastic fibers with few smooth muscle fibers in human spleen while in dog spleen more smooth muscle fibers and few elastic fibers were present. White pulp was more in human than dog. Red pulp and marginal zone of dog spleen showed ellipsoids and penicillar arteries which were poorly defined in human spleen. Red pulp showed prominent venous sinuses in dog spleen than human. **Conclusion:** Histological features are well defined in storage type of spleen as in dog than the defensive human spleen. Histological features of dog spleen are useful in better understanding of microscopic anatomy and can be used for teaching purpose.

Keywords: human spleen, dog spleen, spleen microscopy, comparative spleen histology.

1 Introduction

Spleen is the major secondary lymphatic organ involved in filtering the blood and mounting immune responses against blood borne antigens. The spleen in domestic species has the ability to add to the erythrocyte and granulocyte population and can be a reservoir of red blood cells during periods of unusual demand. (SAMUELSON, 2007) In all mammals the spleen is surrounded by capsule which is covered by peritoneum. The capsule sends trabeculae in the substance of the spleen. The area between the capsule and trabeculae form the splenic pulp which is divided into two parts, hematogenous red pulp and lymphoid white pulp separated by marginal zone. The compartments of splenic white and red pulp in humans are described very differently in primary publications and textbooks of microscopic anatomy. A mixture of findings derived from human, rodent and other experimental animals are described in most textbooks. (STEINIGER, BARTH, HERBST et al., 1997) Spleen has been classified by Banks (1993) in two basic types according to the arrangement of capsular and trabecular smooth muscle fibers. Defensive type has few trabeculae and smooth muscle fibers but abundant lymphatic tissue ex-man, whereas storage type has many trabeculae and smooth muscle fibers. It is relatively large and contains less white pulp ex- horse, dog and cat. Dellman and Brown (2006) classified mammalian spleens depending on the type of post capillary vessels into sinusal and non-sinusal. Among the domestic animals, only dog have typical venous sinuses similar to those of human and rat spleens. Dog spleen is sinusal type and can store large amount of blood for quick release during emergency (BACHA and BACHA, 2000). The present study was aimed at comparing histological differences between human with dog spleen as they differ in functions.

2 Material and methods

The study was undertaken with permission from institutional ethical committee. The spleens of 5 mature dogs were collected from the Department of Physiology, after the department has utilized these animals for their experiment till their death. 5 human spleens were collected from Forensic Medicine Department. Out of these 3 were of deaths due to cardio-vascular disease, 2 were accidental deaths. All subjects were from age group 35-50. Each spleen was perfused with normal saline through splenic artery to remove blood from blood vessels. Later the spleen was infused with 10% buffered formalin through splenic artery and splenic vein was then ligated. The spleen was immersed in Bouin's fluid as a whole for 24 hours to allow it to become harder. Then it was cut into pieces. Paraffin embedding was done. Sections of five microns were cut and mounted on slides. The slides were stained by Haematoxylene and Eosin stain for general histological pattern, Masson's trichrome stain especially for architecture of connective tissue and by Gordon and Sweet's reticulin method for reticular fibers. The slides were labeled and stored for observations under light microscope.

3 Results

3.1 Human spleen

Capsule was thin and consisted of admixture of elastic and collagen fibers with few smooth muscle fibers (Figure 1a). Trabeculae were thick and few in number with more elastic fibers and some collagen fibers (Figure 2a). White Pulp or Malpighian follicle was abundantly scattered throughout the substance of spleen which was in the form of lymphoid nodule traversed by eccentrically placed central arteriole (Figure 3a). Stroma was made up of reticular fibers forming network (Figure 2b). Free cells consisted of compactly arranged small sized lymphocytes. Marginal zone consisted of sinuses and cells such as lymphocytes, macrophages, red blood corpuscles and plasma cells. Small rounded ellipsoids or periarteriolar macrophage sheaths were present in marginal zone. Ellipsoids were identified by the presence of reticular fibers and its macrophage cells around arteriole. Red Pulp was formed by meshwork of reticular fibers. It was permeated by sinuses and cords of cells. Sinuses were elongated and lined with endothelial cells (Figure 4a). Cords were composed of lymphocytes of variable sizes followed by macrophages, red blood corpuscles and plasma cells. Poorly defined ellipsoids were also present (Figure 5a).



Figure 1. a) Human spleen capsule. Masson's trichome stain. $200 \times$ showing collagen and smooth muscle fibers. b) Dog spleen capsule and trabeculae. Masson's trichome stain. $200 \times$ showing smooth muscle fibers. Arrow indicates subcapsular contracted sinuses. Thick arrow indicates trabecula.



Figure 2. a) Human spleen H & E stain. $100 \times$ showing elastic fibers in trabeculae and trabecular blood vessel. b) Human spleen Gorden and Sweet's stain. $200 \times$ showing reticular fibers.



Figure 3. a) Human spleen H & E stain. 200× showing white pulp. b) Dog spleen. Masson's trichome stain. 100× showing white pulp. Arrow indicates ellipsoid in marginal zone.



Figure 4. a) Human spleen H & E stain. 400× showing red pulp. b) Dog spleen. Masson's trichome stain. 200× showing ellipsoids surrounded by sinuses in red pulp. Arrow indicates ellipsoid.



Figure 5. a) Human spleen. H & E stain. 200× showing white pulp, marginal zone and red pulp. Arrow indicates ellipsoids b) Dog spleen Masson's trichome stain. 200× showing sinuses, penicilli and ellipsoids in red pulp. Arrow indicates ellipsoids. Arrowhead indicates penicilli.

3.2 Dog spleen

Capsule was thin with elastic fibers in the periphery. Smooth muscle fibers were abundant and occupy maximum part. Many thick trabeculae containing mainly smooth muscle fibers were seen (Figure 1b). White pulp was abundant with eccentrically placed central arteriole. Stroma was formed by reticular fibers. In marginal zone marginal sinusoids were seen along with macrophages and lymphocytes. Elongated ellipsoids were seen (Figure 3b).

Red Pulp had reticular fibers forming meshwork and had sinuses and cords. It also contained abundant well defined ellipsoids and penicillar arteries (Figure 5b). Elongated and rounded ellipsoids were surrounded by two to three sinuses (Figure 4b). Ellipsoids were randomly scattered throughout the red pulp. Sinuses were lined with flattened endothelial cells. Sinuses were contracted in subcapsular region (Figure 1b). Cords contained lymphocytes of variable sizes, macrophage cells, red blood corpuscles and plasma cells.

4 Discussion

Various functions of spleen include immunological response to antigens, red blood cells production and destruction, blood filtration, storage of blood and phagocytosis. Most histology textbooks depict a mixture of findings derived from human, rodent and other experimental animals. (STEINIGER, BARTH, HERBST et al., 1997). The spleens of different mammals show histological differences according to their functions. The present study was aimed at comparing the histology of human and dog spleen.

4.1 Capsule and trabeculae

In human spleen, the capsule was thin and had elastic and collagen fibers with few smooth muscle fibers. In trabeculae elastic fibers was more than that of capsule matching with the findings of Maximov and Bloom (1975). They observed that human spleen capsule has network of thickest elastic fibers in the deep layers whereas in trabeculae elastic fibers are more than capsule.

In dog spleen capsule was thin. In peripheral part of capsule elastic fibers were present. In inner two third parts, smooth muscle fibers were seen. Aughey and Frye (2001) has described that capsule in dog spleen is relatively thin and consists of elastic and smooth muscle fibers. Dellman and Brown (1981) stated that dog and cat have thinnest splenic capsule and smooth muscle fibers make up more than two third of total thickness of capsule. Dellman and Brown (2006) added to previous findings that capsule consists of two layers: a layer of dense irregular connective tissue and a layer of smooth muscle. Raviola (1994) stated that in animal species in which capsule and trabeculae are rich in smooth muscle fibers, the spleen acts as a store of red blood cells.

We observed few smooth muscle fibers and trabeculae in human spleen where as in dog many smooth muscle fibers and trabeculae were seen. These findings are matching with Banks (1993). He classified the spleen in two basic types according to the arrangement of capsular and trabecular smooth muscle fibers. Defensive type has few trabeculae and smooth muscle fibers as in human. Storage type has many trabeculae and smooth muscle fibers as seen in horse, dog and cat. Intermediate forms are typical of ruminants and swine. According to Ham and Cormack (1979) in some animals, there are much more smooth muscles in capsule than in man and its contraction can materially assist the smooth muscles of trabeculae in contracting the spleen and so forcing the blood it contains into system at times of emergency.

4.2 White pulp

We observed abundant lymphoid tissue in both spleens. Comparatively dog spleen had less white pulp than human spleen. Banks (1993) found abundant lymphoid tissue in human and classified as defensive type. They classified spleen with less amount of white pulp as storage type which is seen in dog. Raviola (1994) observed that white pulp of mammalian spleen is composed of the periarterial lymphatic sheath and lymph follicles. Steiniger, Barth, Herbst et al. (1997) found that lymph follicles predominate in human spleen.

4.3 Marginal zone

In human and dog spleen we observed sinusoids and cells such as lymphocytes, macrophages, red blood corpuscles, plasma cells. Elongated ellipsoids were seen in dog. Jacobson (1971) studied ellipsoids of dog and found that ellipsoids of marginal zone encircle the white pulp nodule and are more elongated than red pulp ellipsoids. Dellman and Brown (1981) stated that in pig, ellipsoids are larger and abundant and a many are located at marginal zone. Their location is similar in cat, dog and horse but less numerous and smaller than those of pig. Our findings about ellipsoids in dog are nearly matching to above studies. Though small sinuses were present in marginal zone, marginal sinus was totally lacking in both. Steiniger, Barth, Herbst et al. (1997) stated that in human spleen perifollicular zone is present around white pulp. Macrophage sheaths occur primarily in this zone and they are rather small structures. We observed similar findings.

4.4 Red pulp

Apart from cords and sinuses, red pulp of dog contains abundant ellipsoids and penicillar arteries. Ellipsoids are often surrounded by two to three sinuses. Rounded and elongated ellipsoids are randomly scattered throughout the red pulp. Our findings correlate with Lewis (1957). They observed that sinusoids are better developed in sheep and more in dog and human spleen. In dog, sinusoids are related to ellipsoids. Dellman and Brown (1981) stated that spleens of domestic animals are sometimes classified according to blood storage capacities. The spleens of horse, dog and pig have abundant venous sinuses and are said to be sinusal type. Therefore, they store large amount of blood for quick release during emergency. Jacobson (1971) studied splenic ellipsoids and observed that in dog, ellipsoids are found in the red pulp and marginal zone but never in malpighian follicles. Ellipsoids are randomly scattered. Their shape is also dependant upon the variations in the number and branching of axial capillaries. Dog ellipsoids are basically spherical or elliptical. Sinuses encircle the ellipsoids. Such findings are also seen in our study.

Ham and Cormack (1979) stated that ellipsoids are developed to different degrees in different species. Those of man are not highly developed. Ellipsoids represent nothing more than a condensation of reticular fibers and reticuloendothelial cells around a capillary. Ellipsoids seem to be highly developed in those species in which capsule and trabeculae contain more amounts of smooth muscle fibers. In these animals, it is not unlikely that the ellipsoids cut down the amount of blood entering the spleen, when the smooth muscle of the capsule and trabeculae contract in emergency and certain other conditions. It is doubtful that more or less rudimentary ellipsoids in man act to any extent even as passive sphincter. They have openings between their reticuloendothelial cells through which the blood from the central capillary can escape. They serve as the first filters that arterial blood encounters in the spleen. Ellipsoids were poorly defined in human spleen. They are rounded and surrounded by few macrophages. Kelly, Wood and Enders (1985) observed that sheaths are not prominent in human as in dog. Steiniger, Barth, Herbst et al. (1997) stated that ellipsoids do not occur in human and the impression that sheathed capillaries are also present in the red pulp is probably due to tangential sections of perifollicular areas which are difficult to distinguish as such.

5 Conclusion

Storage type of spleen like dog show well defined histological features than defensive type of spleen like human. Histological features of dog spleen are useful in better understanding of microscopic anatomy and the slides can be used for teaching purpose in addition to human spleen.

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