Indian Journal of Veterinary Anatomy 38 (01):36-39, June 2026

Micromorphological Studies on Hemal Nodes of Sheep

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Received: 06.02.2026; Accepted: 20.06.2026

ABSTRACT

The present study was conducted on tissue samples from hemal nodes of apparently healthy adult sheep (n=6) collected from slaughter house and postmortem hall to elucidate its micromorphological features. Micromorphological observation revealed that hemal nodes were surrounded by capsule. A few connective tissue trabeculae extended from the capsule into the parenchyma. Underneath the capsule and trabeculae, subcapsular and trabecular sinuses were present. The subcapsular and trabecular sinuses were filled with erythrocytes with few lymphocytes. Both primary and secondary lymphoid follicles were randomly distributed in the parenchyma of hemal nodes. There was no clear demarcation between cortex and medulla. The medulla comprised of medullary cords and sinuses. The cellular elements observed in the parenchyma of hemal nodes were lymphocytes, macrophages, plasma cells, and megakaryocytes. It was concluded that hemal nodes of sheep exhibited a capsule with trabeculae, sinuses filled mainly with erythrocytes, randomly distributed lymphoid follicles without cortex–medulla distinction, and parenchyma composed of lymphocytes, macrophages, plasma cells, and megakaryocytes.

Keywords: Hemal nodes, Histomorphology, Sheep.

INTRODUCTION

Hemal nodes are unique structures in various mammals, including ruminants. In ruminants, hemal nodes are distributed in different body regions viz., neck and mesenteric regions along large blood vessels in thoracic and abdominal cavities and subcutaneous region of head (Ezeasor and Singh, 1988). Hemal nodes of sheep are lymphoid organs within the blood circulation encased by connective tissue capsule. The proposed functions of hemal node include erythropoiesis, platelet formation, blood storage and immune cell activation (Cerutti and Guerrero, 2008). However, still the precise functions of hemal nodes have not been clearly established. The elucidation of histomorphological features of hemal nodes in sheep in the present study will help establish the possible functions of hemal node in sheep.

MATERIALS AND METHODS

The present study was conducted on tissue samples from hemal nodes of apparently healthy adult sheep (n=6) collected from slaughter house and postmortem hall. Tissue sample collected from different regions viz., mesenteric and along large blood vessels**.** To study the histomorphology of hemal node of sheep, tissue samples were fixed in ten percent neutral buffered formalin (NBF) and Bouin's fluid. After fixation was achieved, the tissue samples were processed for paraffin blocks preparation by acetone-benzene schedule (Luna 1968) and sections of 3-5 μm were obtained on glass slides with rotary microtome. The paraffin sections were stained with various stains for the micromorphological study like hematoxylin and eosin for routine morphology, Masson trichrome for collagen fibres (Luna, 1968). Gridley for reticular fibres, (Sheehan and Hrapchak, 1973), Verhoeff's for elastic fibres (Sheehan and Hrapchak, 1973).

RESULTS AND DISCUSSION

The hemal nodes of sheep were surrounded by a connective tissue capsule (Fig. 4). The capsule was composed of connective tissue fibers, mainly reticular and collagen fibers (Figs 4 & 6). The

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adipose tissue was observed inside the capsule at many locations (Fig. 6). From the capsule, trabeculae extended into the parenchyma of hemal node containing reticular and collagen fibers. Gargiulo *et al.* (1987) reported similar observations in hemal nodes of sheep. Zidan *et al* (2012) also observed connective tissue capsule and trabeculae comprising collagen and reticular fibers in hemal nodes of buffalo. The capsule was composed of collagen and reticular fibres with a connective tissue framework (Fig 2, 4).

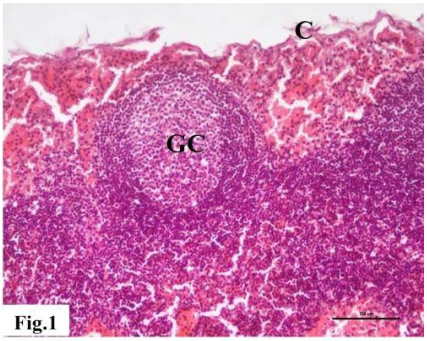
Below the capsule, the subscapular sinus was present; however, in the present study, a uniformly well-differentiated subcapsular sinus was not observed in the hemal node of sheep (Figs. 1 & 5). In contrast, Sur *et al* (2005) reported that the sub-capsular sinus was developed in the capsule of the sheep hemal nodes. Ozaydin *et. al.* (2012) also reported presence of sub-capsular sinus below the capsule in goat hemal node. Both the subcapsular and trabecular sinuses were lined by endothelial cells and filled with erythrocytes, with occasional lymphocytes (Fig. 3 & 11), in agreement with the reports of Sur *et al.* (2005) in sheep.

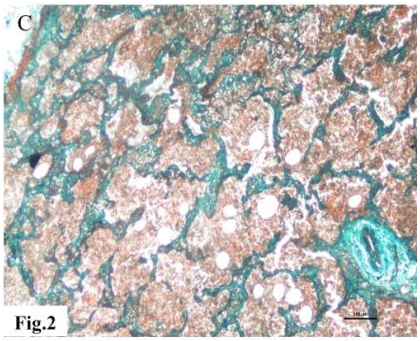
The parenchyma of sheep hemal node was comprised of cortex and medulla which were intermingled and lacked a clear demarcation (Fig.11). Similar findings have been reported by Gargiulo *et al* (1987) in sheep. Conversely, Zidan and Pabst (2004) reported that the parenchyma of the sheep hemal node is clearly differentiated into an outer cortex and an inner medulla. Kaur *et. al.* (2018) also reported well-demarcated cortex, paracortex, and medullary regions in the hemal nodes of buffalo. The connective tissue support of the parenchyma was mainly contributed by reticular fibres forming a framework (Fig.6). Similar observations were recorded in the hemal nodes of goat (Ezeasor and Singh, 1988). Kaur *et. al.* (2018) also reported that supportive network of parenchyma was mainly contributed by reticular fibres in buffalo

The cortical region of the hemal node was comprised of primary and secondary lymphoid follicles with lightly staining germinal centre (Figs. 8 & 10). The majority of lymphoid follicles especially in the area of subcapsular sinus were secondary lymphoid follicles with light staining germinal centre (Fig. 11). The cellular elements of hemal node of sheep parenchyma contained lymphocytes, erythrocytes, macrophages, plasma cells and megakaryocyte (Figs. 9 & 10).The presence of cellular elements indicated the involvement of the hemal nodes in the erythropoiesis, erythrophagocytosis, and immune functions. Sur *et al.* (2005) also reported lymphatic nodules with germinal centers and lymphatic cord in sheep. These were localized in the germinal centres of secondary lymphatic nodules and in lymphatic cords. Similar reports have been mentioned by Kaur *et. al.* (2018) in buffalo.

The medullary region of the hemal node was comprised of medullary cords containing lymphocytes and medullary sinuses filled with mainly erythrocytes (Figs. 8,10,11). Zaghloul & Derbalah (2014) observed macrophages engulfing erythrocytes in Egyptian water buffalo. Plasma cells were found in the vicinity of macrophages. This suggested the role of plasma cells in helping macrophages perform erythrophagocytosis in hemal nodes, in agreement with the reports of Kaur *et al.* (2019).

Both the capsule and parenchyma in sheep hemal nodes were enriched by blood vessels (Figs 3,4,7), however, lymph vessels could not be localized. Park *et.al.* (2001) also made similar observations in the hemal nodes of sheep. However, Ezeasor and Singh (1998) and Ozaydin *et. al.* (2012) observed both blood and lymph vessels in the parenchyma of goat hemal node. Choudhary *et. al.* (2018) also observed lymph vessels in goat hemal nodes.





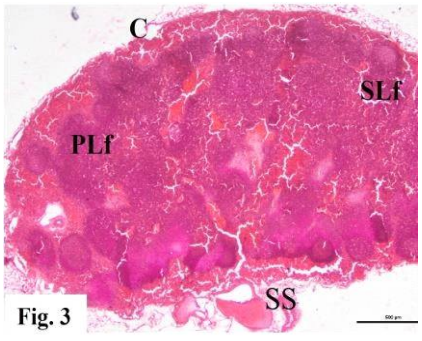
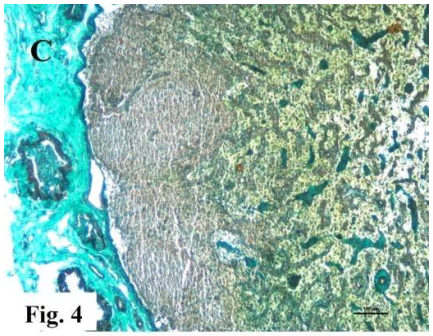
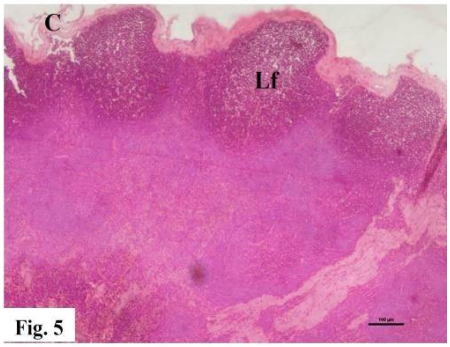


Fig.1. Photomicrograph showing the the capsule(C), germinal centre(GC) in the hemal node of sheep. H& E X20, **Fig.2**.Photomicrograph showing capsule(C)and trabeculae having collagen fibres. Masson's trichome X 4., **Fig.3**.Photomicrograph showing the capsule (C), primary and secondary follicles (Plf, Slf), medullary cords and sinusoids (SS) in the hemal nodes of sheep. H & E X 4





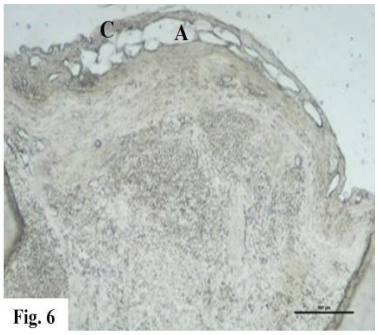
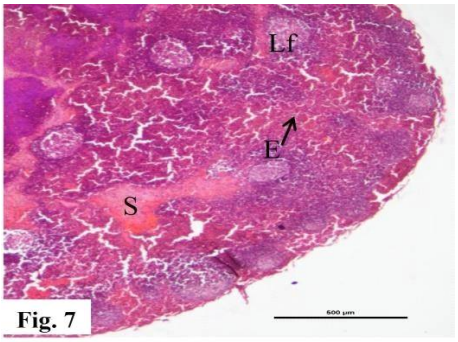
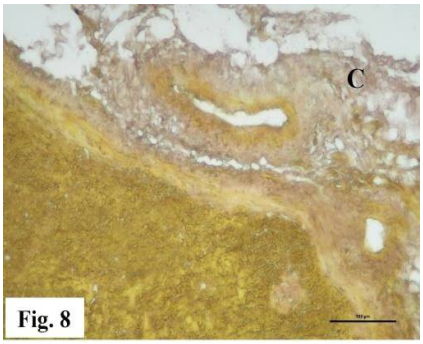


Fig.4. Photomicrograph showing the capsule (C) and trabeculae with collagen fibres in the hemal nodes of sheep. Masson's trichome X 4. **Fig5.** Photomicrograph showing the presence of capsule (C), secondary lymphoid follicle (Lf), underneath the capsule in the hemal nodes of sheep. H&E X 10, **Fig 6.** Photomicrograph showing the reticular fibres in parenchyma and capsule (C). Adipose tissue (A) at locationsin capsule. GridleyX20.





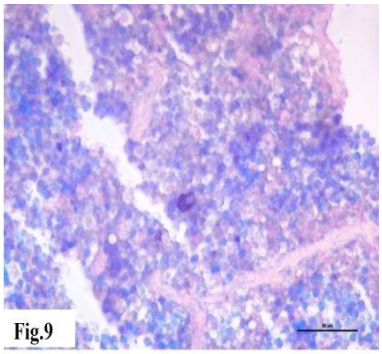
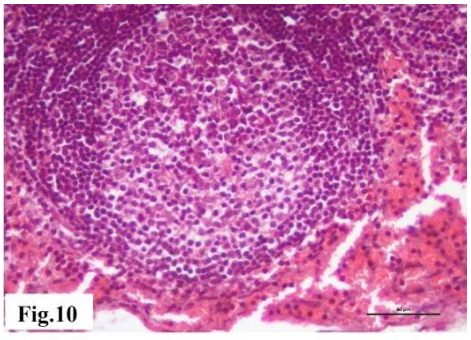
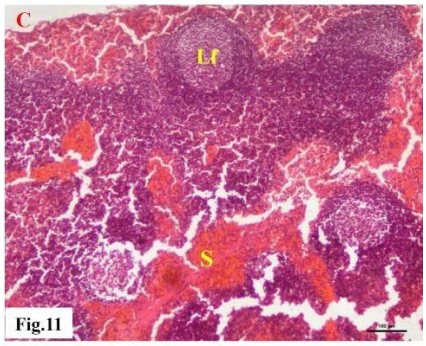


Fig.7. Photomicrograph showing the presence of lymphoid follicles (Lf), medullary cords and sinusoida (S) filled with erythrocytes in hemal nodes of sheep. H&E X 4., **Fig.8.** Photomicrograph show the presence of elastic fibres in the blood vessels of capsule (C). Verhoeff X20., **Fig.9.** Photomicrograph showing the presence of megakaryocytes (Arrow) in hemal nodes of sheep. GiemsaX 40





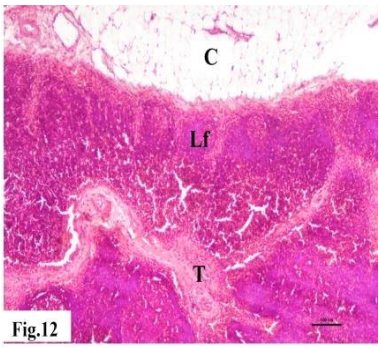


Fig.10. Photomicrograph showing the presence of Plasma cells (P) in hemal node of sheep. H&E X 40. **Fig.11**. Photomicrograph showing the presence of lymphoid follicle(Lf), Capsule (C), Sinusoids(S) with the erythrocytes in hemal node of sheep. H&E X 10 **Fig.12.** Photomicrograph showing the presence of lymphoid follicle(Lf), Capsule (C), with adipose tissue and trabeculae in the hemal nodes of sheep in hemal node of sheep. H&E X 10,

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