

HISTOMORPHOLOGICAL, HISTOCHEMICAL AND HISTOENZYMIC STUDIES ON LYMPHOID TISSUE OF CAECUM IN KHAKI CAMPBELL BREED OF DUCK

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Received: 29.19.2021; Accepted: 27.11.2021

SUMMARY

Caecum is the part of gut associated lymphoid tissue and play important role in immune system. The present work was carried out on lymphoid tissue of caecum in the Khaki Campbell breed of duck at various age groups in 30 birds of either sex. In the caecum, the diffuse lymphatic tissue was observed in the lamina propria in Group-I and Group-II. The lymphoepithelium was containing intra- epithelial lymphocytes and goblet cells. In Group- III, the lymphatic nodules were found adherent to the tunica muscularis and were covered by capsule of delicate collagen fibers. In Group- IV, the aggregated form of lymphatic tissue was occurred between the core of muscularis mucosa. In caecum, the lymphatic nodules showed the strong PAS positive reaction in Group-II, Group-III and Group-IV. The PAS-AB reaction was weak to moderate in lymphatic nodules and lymphoepithelium of in all groups. The caecal tonsils showed strong Alkaline phosphatase reaction. The epithelium depicted mild Alkaline phosphatase activity. Caecal tonsils showed strong Acid phosphatase reaction in the lymphatic nodules.

Keywords: Caecum, Duck, Histology, Lymphoid tissue

How to cite: Gedam, P.M. and Nandeshwar, N.C. (2022). Histomorphological, histochemical and histoenzymic studies on lymphoid tissue of caecum in khaki campbell breed of duck. *Haryana Vet.* 61(SI-2): 138-140.

The gut associated lymphoid tissue (GALT) plays a vital role in the immune system by protecting the mucosa against the harmful antigens that enter the body through food and air. The scanning of the literature shows that the GALT is studied in avian species and it is distributed throughout the gastrointestinal tract as pharyngeal tonsil (Crole and Soley, 2012), oesophageal tonsil (Olah *et al.*, 2003 and Nagy *et al.*, 2005), pyloric tonsil (Nagy and Olah, 2007) and caecal tonsil (Kitagawa *et al.*, 1998). The ducks, as they reared in rural area as backyard farming, might get exposed with many pathogens. It is therefore necessary to know how gut wall respond to these antigens and acts as major line of defence mechanism. Hence, the present study on lymphoid tissue of caecum in Khaki Campbell breed of duck was undertaken.

The present work was carried out on lymphoid tissue of caecum in the Khaki Campbell breed of duck at various age groups in 30 birds of either sex. These ducks were divided in to 5 age groups comprising 6 birds in each group. Group-I (0-2 weeks), Group-II (2-4 weeks), Group-III (4-6 weeks), Group IV (6-12 weeks), Group V (12-18 weeks). The tissue samples after collection were washed with normal saline and then subjected for fixation. The tissue pieces were fixed in 10% neutral buffered formalin solution and Bouins fluid and processed for paraffin technique as per the method of Drury and Wallington (1980). The sections of 5-6 μ thickness were obtained on rotary microtome and stained for histomorphology, histochemistry and histoenzymology staining procedures.

Haematoxylin and Eosin staining for general histomorphological observations, Masson's trichrome staining method for demonstration of collagen and smooth muscles fibers. (Singh and Sulochana, 1996). Gomori's stain for reticular fibers, Verhoeff's stain for elastic fibers, Periodic Acid Schiff's reaction for presence glycogen, PAS-AB method for both neutral and acid mucopolysaccharides, Gomori alkaline phosphatase method for alkaline phosphatase activity and Gomori acid phosphatase method for acid phosphatase activity (Bancroft and Stevens, 1982). The observation on the histomorphology, histochemistry and histoenzymology were recorded and the photomicrographs were taken in Nikon 80 i microscope.

Histomorphology: In the caecum the diffuse lymphatic tissue was distributed in the mucosa of body and terminal part, but aggregated lymphatic nodules were occurred in the proximal part (neck) near the ileo-caecal junction. The characteristic caecal tonsil were not observed in the Khaki Campbell duck. These finding were in accordance with Georgescu *et al.* (2007) and Senapati *et al.* (2015). Nickel *et al.* (1977), stated that, there was a reduction of lymphoreticular tissue towards cupola shaped end of the caeca in duck and goose.

In Group-I and Group-II, the diffuse lymphatic tissue was observed in the lamina propria, the lymphatic nodules were also observed in the sub-mucosa (Fig. 2). The lymphoepithelium was comprised of intra epithelial lymphocytes and goblet cells. In Gr. III, the lymphatic nodules were observed adherent to the tunica muscularis

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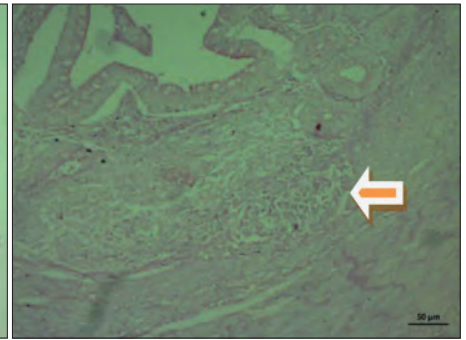
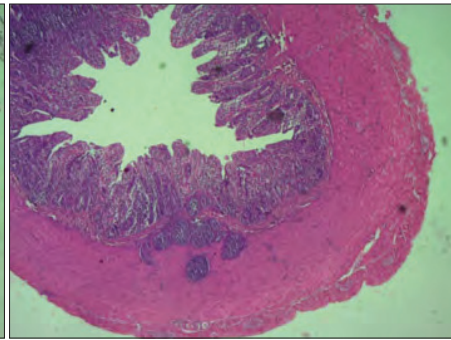
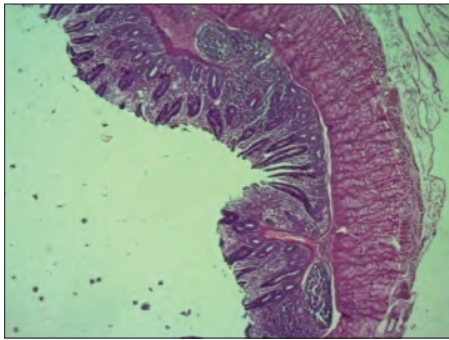


Fig. 1. Photomicrograph of proximal caecum (Group-III) showing, A) Diffuse lymphatic tissue, B) Lymphatic nodule, C) Mucous glands (H.E. 50X)

Fig. 2. Photomicrograph of proximal caecum (Group-II) showing, A) Diffuse lymphatic tissue; B) Aggregated lymphatic nodules in muscularis externa. (H.E. 50X)

Fig. 3. Photomicrograph of proximal caecum (Group-III) showing PAS reaction in lymphatic nodule. (PAS 200X)

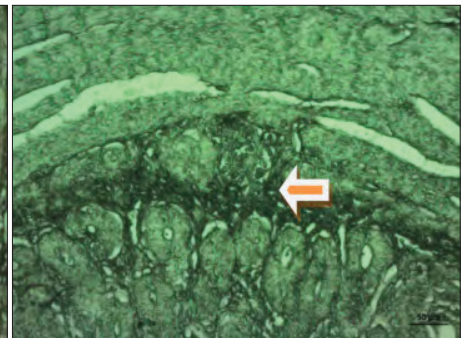
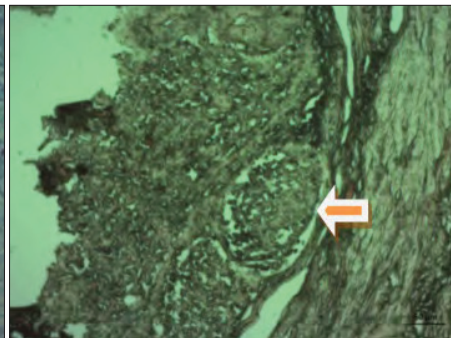
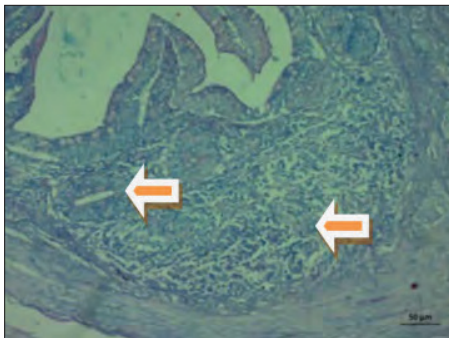


Fig. 4. Photomicrograph of proximal caecum (Group-IV) showing PAS-AB reaction in lymphatic nodule and mucous gland. (PAS-AB 200X)

Fig. 5. Photomicrograph of proximal caecum (Group-IV) showing Alkaline phosphatase reaction in lymphatic nodule. (Alkaline phosphatase 200X)

Fig. 6. Photomicrograph of proximal caecum (Group-IV) showing Acid phosphatase reaction in lymphatic nodule. (Acid phosphatase 200X)

and were covered with delicate collagen fibers capsule, but some area of nodules was devoid of capsule and were communication with the follicular lymphatic tissue of the lamina propria (Fig. 1). In Group-IV, the aggregated form of lymphatic tissue was observed between the core of muscularis mucosa. In Group-V, the mucosa was infiltrated with the diffuse lymphatic tissue. Lymphatic nodules were also observed in the tunica muscularis. These nodules were separated from each other with inter follicular connective tissue (Kitagawa *et al.*, 1998). Many capillaries were also occurred in the inter-follicular areas. The epithelium covering the follicle was consisted of intraepithelial lymphocytes and goblet cells. Proliferation of the epithelium was also observed between the follicles.

The present study records the lymphatic tissue in lamina propria, sub-mucosa and also in muscularis externa. The infiltration was more in proximal part of the caecum. These results corroborates with Kitamura *et al.* (1976) and Georgescu *et al.* (2007) in duck. According to the Usha Kumary *et al.* (2002), the caecal tonsils were occurred in the middle and distal part of the caecum in Japanese quail. Kitagawa *et al.* (1998) stated that the aggregated lymphatic nodules in the different region of the caeca of chicken could provide the immunological

surveillance against caecal luminal context.

Histochemistry: In proximal caecum the lymphatic nodules showed the strong PAS positive reaction in Group-II, Group-III and Group-IV (Fig. 3). The PAS positive reaction was moderate in lymphatic nodules of caecum in Group-I and Group-V. The PAS-AB reaction was weak to moderate in lymphatic nodules and lymphoepithelium of caecum in all groups (Fig. 4).

Histoenzymology:

Alkaline phosphatase: The caecal tonsils showed strong activity in Group-III, IV and Group-V (Fig. 5). The epithelium depicted mild activity Group-III, IV and Group-V (Fig. 5). Raju *et al.* (2012) stated that follicle associated epithelium of the Peyer's patches of sheep showed positive activity for alkaline phosphatase which was more intense over the follicle domes. Burns and Maxwell, (1986) suggested that, the faults in PP epithelium, was positive for alkaline phosphatase and allowed the extrusion of lymphocyte into the intestinal lumen. According to Gautam *et al.* (2014), the surface epithelium of small intestine showed moderate activity in caprine gut. They also reported positive reaction in lymphatic nodules for T cell area and the neck portion of

the lymphoid follicle.

Acid phosphatase: Caecal tonsils showed strong activity in the lymphatic nodules (Fig. 6). Raju *et al.* (2012) observed an intense enzyme activity in the follicle associated epithelium and dome areas of ileal Peyer's patches in sheep. Gautam *et al.* (2014) found that, the lymphoid follicles of both Peyer's patches and solitary lymphatic nodules were positive for acid phosphatase activity in the caprine.

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