



AGE RELATED CHANGES IN THE HISTOLOGY OF THE BURSA OF THE DOMESTIC FOWL (*GALLUS DOMESTICUS*)*

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Abstract:

Marked phagocytic activity was seen in day old stage. Interfollicular and follicular epithelium varied in staining reactions. Regression started at 8 weeks and completed by 24 weeks. During involution cysts, epithelialisation of follicles, dendritic cells were conspicuous.

Keywords: *Involution, Giriraja, bursa, histology.*

The bursa of fabricius is a lympho-epithelial gland unique to class aves and is located dorsal to the cloaca (Hodges, 1974). It is a primary lymphoid organ where B cells differentiate and participate in humoral response. While elaborate studies have been undertaken on immune system of chicken, not much work has been done on age related changes in the lymphoid organs of Giriraja birds. Giriraja is a disease resistant bird with body weight and egg production three times that of local bird. The present communication deals with the histology and age related changes in the bursa of fabricius of Giriraja birds.

Materials and Methods

A total of 72 birds were reared separately at the UAS poultry farm, Bangalore from day old to 24 weeks and the bursa was collected from six birds each every alternate week. The materials collected were fixed in 10% neutral buffered formalin, Bouin's fluid, lymphatic tissue fixative and Zenker's acetic fluid. The tissue pieces were processed for paraffin embedding. Sections were cut at 4 mm thickness and were stained by Hematoxylin and eosin staining, Van Geison's stain for collagen fibres, Masson's Trichrome method, method for reticulin, Weigert's elastic stain, Methyl green Pyronin method for the demonstration of DNA and RNA.

Results and Discussion

The wall of the mature bursa was composed of three tunics, the tunica mucosa, tunica muscularis and tunica serosa. The mucosa was lined by pseudostratified epithelium and presented polyhedral follicles closely packed together with small amounts of connective tissue in the lamina propria. This observation is in accordance with that of Hodges (1974).

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The follicles in day-old birds were mainly of two kinds namely, projectile follicles and non-projectile follicles. It possessed more number of lymphoblasts than lymphocytes. Histiocytes surrounded by empty spaces giving a starry sky appearance was an important feature of this age group. Similar reports were made earlier by Freeman (1984). These cells possessed remnants of old dying cells in various stages.

In the first few weeks, the connective tissue increased considerably in relation to the follicles and the bursa became fibromuscular with age. This concurs with the observations of Yamada *et al.* (1977).

Epithelium lining the folds was pseudostratified columnar while it was



Fig. 1. Basement membrane of corticomedullary junction showing positive reaction for reticular stain

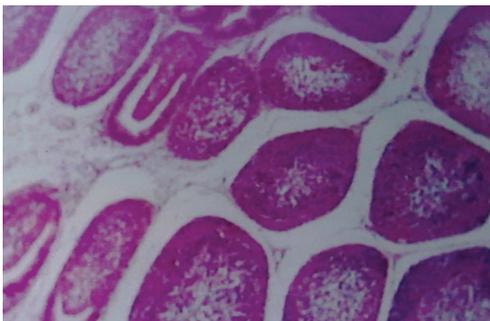


Fig. 2. Elongated dendritic cells in the involuting bursal follicles

simple columnar in the crypts similar to the observations of Malewitz and Calhoun (1958) in fowl. At least three rows of nuclei were visible in the epithelium. The superficial row was elongated, oval and deeply basophilic with clumped chromatin. The middle row of nuclei was larger, oval and pale staining with prominent single nucleolus. The basal or

deep row of nuclei presented oval pale staining nucleus with prominent single nucleolus. The cytoplasm of these cells presented supranuclear vacuolations. The epithelial tuft cells present at the tip of the follicle, were low columnar with pale cytoplasm. These observations concurred with earlier reports of Hodges (1974) in the fowl.

The interfollicular epithelium (IFE) released mucin into the bursal lumen, as indicated by the alcian blue positive reaction. This reaction however was absent in the FAE (Follicle associated epithelium). Vacuoles were observed in the follicle-associated epithelial cells in accordance with the reports of Bockman and Cooper (1973).

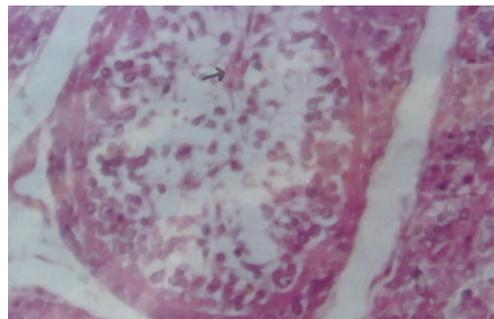


Fig. 3. Epithelialisation of bursal follicles during involution.

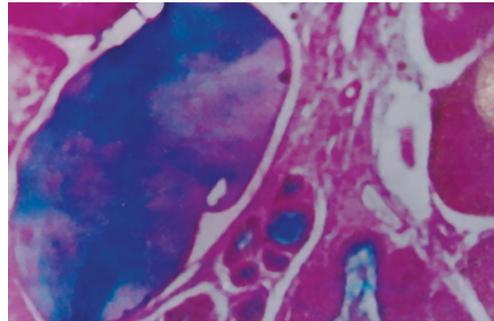


Fig. 4. Cysts in involuting bursa positive for alcian blue reaction.

As reported by Farner *et al.* (1983) the bursal follicle was divided into a medullary FAE unit and an extramedullary unit, comprising the cortex. The medulla was separated from the bursal lumen by FAE.

Rodak *et al.* (1969) stated that the cortex of one week old chicken was distinct

but in the present study the cortex and medulla were clearly differentiated only by four weeks. After 5 weeks a basophilic cortex and an eosinophilic medulla, characterized by paucity of lymphocytes were evident. A dense network of capillaries separated the cortex from the medulla surrounded by a basement membrane. A layer of pale staining reticuloepithelial cells formed the bulk of the corticomedullary junction similar to the observations of Scala *et al.* (1989) in duck (Fig.1).

The cortex stained more deeply than the medulla due to its higher concentration of small lymphocytes. Lymphoblasts, lymphocytes, macrophages containing nuclear remnants and plasma cells we seen in it.

The medulla contained reticular cells, lymphoblasts, lymphocytes, macrophages and plasma cells. Farner *et al.* (1983) made similar observations in chicken.

A distinct bursal follicular reticular cell and a medullary epithelial structure resembling Hassal's corpuscle of thymus were also identified. These findings give further support to the idea that the avian bursa has functions of a peripheral lymphoid organ as well as containing T lymphocytes. (Naukkarinen, 1982)

The reticuloepithelial cells formed the supportive framework of both the cortex and medulla in the meshes of which lymphocytes were present. The dendritic cells were also noted in addition to the reticuloepithelial cells, as per the report of Dourain *et al.* (1976) (Fig. 2). Plasma cells were located in the interfollicular tissue and beneath the bursal epithelium in addition to the follicles.

The bursa of Fabricius started regressing at the age of eight weeks post hatch contrary to the findings of Millicevic *et al.* (1986) who noted that involution started at 24 weeks.

At this age, eosinophilic, elongated and flask-shaped follicles of various size were evident. Epithelial canalization of follicles (Fig. 3), lymphocyte depletion and replacement by fibroblasts and adipose tissue, extension of follicular medulla, and accumulation of eosinophilic aggregates containing degenerating lymphocytes with pyknotic nucleus were also seen.

An increased convolutions of the IFE observed during involution was also noted by Farner *et al.* (1983). Alcian blue positive simple tubular glands lined by columnar cells were noticed from 10 weeks of age closely associated with the cysts (Fig. 4). These findings concur with those observed by Scala *et al.* (1989) in ducks. Goblet cells were also seen during involution. Epithelial proliferation followed by degradation and exfoliation was noticed by the 12th week. Vacuolation of the epithelial cells was noticed which increased.

Large cysts lined by simple squamous epithelium enclosing a central core of mucoid substances were found in the medulla at eight weeks and their number increased with age. Initially the cysts were organized with simple squamous epithelium. In the terminal involuted bursa the cysts were surrounded by thin rim of lymphoid cells interposed in dense connective tissue. Free lymphocytes or lymphoid aggregates were seen in the lumen in early stages of bursal involution. Changes in plical or interplical epithelium apparently allowed spillage of lymphocytes into the lumen from degenerating follicles. Areas of epithelial attenuation, vacuolation and folding and detachment were noted over the plical surface and in the interplical areas especially numerous in the interplical areas but were also noted over the plical surface. At 20 weeks of age, the plical epithelium could not be identified. These findings were similar to the observations made by Bickford *et al.* (1985).

With the loss of lymphocytes from the medulla, the FAE dipped into the follicle giving a button - like appearance. The connective tissue stroma proliferated and the macrophages infiltrated the necrotic area of the follicles which were also recorded by Bickford *et al.* (1985) in the fowl.

During involution loss of lymphocytes from the cortex and medulla gave an acinar structure to the follicle. Naukkarinen (1982) stated that the post involutory bursa was devoid of lymphoepithelial structures, but in the present study some lymphoid follicles similar to embryonic follicles were found even in the involuted bursa. Large isolated lymphoid follicles lying in the vicinity of small completely retrograde follicles were seen.

The structure of the involuted bursa was similar to that described by Riddel (1987) consisting of fibrous tissue, disorganized smooth muscle, isolated lymphoid follicles and macrophages. By the end of 24 weeks, hyalinization of collagen fibres featured by homogenous, eosinophilic glassy appearance of collagen with sparse cellularity interposed with adipose tissue was seen.

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