

AGE RELATED HISTOCHEMICAL STATUS OF BURSA OF FABRICIUS IN JAPANESE QUAIL (*Coturnix coturnix japonica*)

S.D. Kadam^{1*} and J.Y. Waghaye²

Department of Veterinary Anatomy and Histology,
College of Veterinary and Animal Sciences, Parbhani
Maharashtra Animal and Fishery Sciences University, Nagpur, Maharashtra

ABSTRACT

Histochemical study was conducted on Bursa of Fabricius of 48 normal Japanese quail birds up to four weeks of age, which were divided into four groups of 12 birds each, at the end of first week, second week, third week and fourth week of age, irrespective of sex. In all age groups, moderate to high periodic acid Schiff's (PAS) activity was observed in the interfollicular connective tissue septa. Weak PAS activity was observed in the cortex in all age group of birds. The surface epithelium showed moderate PAS activity almost in all age group of birds, along with high PAS activity in the apical border of the surface epithelium in group I age of birds which was intense from group II age of birds. High Alcian Blue Periodic Acid Schiff's (ABPAS) activity was found in the interfollicular connective tissue septa in group I birds which gradually decreased with the advancement of age. High to intense ABPAS activity was observed in the capillaries as well as tunica intima and adventitia of blood vessels in birds of all age groups.

Keywords: Japanese quail, Histochemistry, PAS, ABPAS, Bursa of Fabricius

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INTRODUCTION

Immune system comprises primary and secondary lymphatic organs. The bursa of Fabricius is the primary lymphoid organ in avians. It is accountable for humoral immunity and plays an important role in maturation of B lymphocytes (Glick, 1956). The bursa

of Fabricius in quail exhibits structural modifications and atrophies as age progresses (Sonfada *et al.* 2014). The purpose of the current study was to better understand the histochemical changes that occur in the bursa of Fabricius with ageing and its significance.

MATERIALS AND METHODS

The present study was conducted in the Department of Veterinary Anatomy and Histology, College of Veterinary and Animal

¹Laboratory Technician, Corresponding author E-mail
id:shrikant430@gmail.com

²Associate Professor

Sciences, Parbhani. The present study was conducted in 48 Japanese quail birds (*Coturnix coturnix japonica*) irrespective of sex reared in the poultry farm of College of Veterinary and Animal Sciences, Parbhani under standard managemental quail rearing practices. They were divided into four groups of 12 birds each, at the end of first week, second week, third week and fourth week of age. The birds were sacrificed by cranial subluxation and the bursa of Fabricius was collected from the dorsal side of cloaca by excising the large intestine.

The collected specimens were washed with normal saline and fixed in 10% neutral buffered formalin, 10 % formal saline and Bouin's fluid. Then the tissue was processed for routine paraffin embedding as per the method of Drury and Wallington (1980). Sections of 5 µm thickness were stained using following staining procedures for histochemical studies:

1. Periodic acid Schiff's method for glycogen (Singh and Sulochana, 1996)
2. Alcian blue periodic acid Schiff's method for acid mucopolysaccharides (Drury and Wallington, 1980)

RESULTS AND DISCUSSION

In the present study, moderate to high Periodic acid Schiff's activity for the presence of glycogen was noticed in the interfollicular connective tissue septa and surface epithelium in all age group of birds (Fig 1, 2, 3 and 4). However, the PAS activity that was observed in the apical border of the surface epithelium in group I birds was intense from group II birds onwards. The apical part of surface epithelial cells showed PAS positive cytoplasmic granules in all age group of birds and were

more intensely stained in group III and IV birds (Fig 1, 2, 3 and 4).

The cortex in all age group of birds demonstrated only weak PAS activity. PAS activity was absent in the medulla of group I birds, whereas it was high from group II birds onwards. However, the extent of PAS activity was observed to be reduced gradually in the medulla from group III birds. The cortico-medullary border showed moderate to high activity in all age group of birds except group I (Fig 1, 2, 3 and 4). In the present study, tunica muscularis showed weak to moderate PAS activity, whereas high to intense activity was observed in all the bursal capillaries as well as tunica intima and adventitia of blood vessels in all age group of birds.

In agreement with the present observations, Jain *et al.* (2010) in poultry birds and Leena *et al.* (2012) in fowl reported PAS activity in the surface epithelium, bursal outer wall and corticomedullary border. Gulmez and Aslan (1999) reported granulated PAS positive epithelial cells in Native Geese. They further noted the group of PAS positive stained cells in the centre of the follicles. The observations of the present study also agree with the reports of Singh *et al.* (2019) in Chabro bird. They mentioned increase in the intensity of PAS positive reaction in granules of surface epithelial cells with age.

The presence of acid mucopolysaccharides was identified by using the Alcian Blue Periodic Acid Schiff's (ABPAS) method. During the present study, high ABPAS activity was found in the interfollicular connective tissue septa in group I birds which was found to be gradually decreased with the advancement of age (Fig

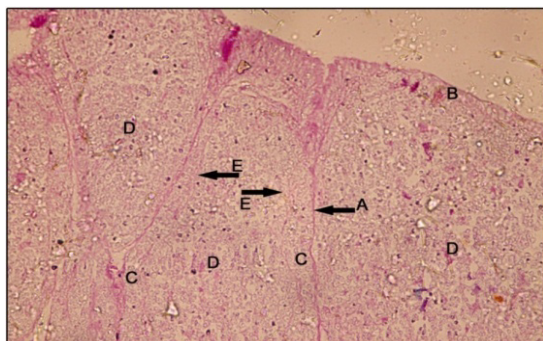


Figure 1: Photomicrograph of bursa of Fabricius in 1 week old Japanese quail

- A. Interfollicular septa
 - B. Surface epithelium
 - C. Cortex
 - D. Medulla
 - E. Corticomedullary border
- (Periodic acid Schiff's, X400)

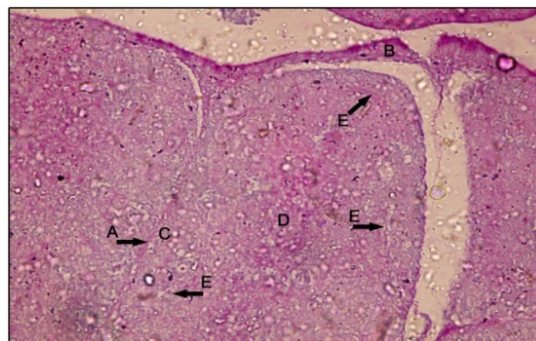


Figure 3: Photomicrograph of bursa of Fabricius in 3 week old Japanese quail

- A. Interfollicular septa
 - B. Surface epithelium
 - C. Cortex
 - D. Medulla
 - E. Corticomedullary border
- (Periodic acid Schiff's, X400)

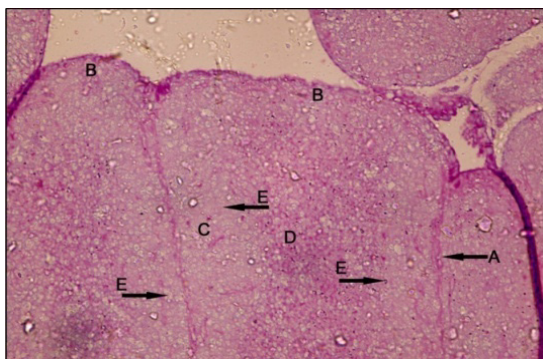


Figure 2: Photomicrograph of bursa of Fabricius in 2 week old Japanese quail

- A. Interfollicular septa
 - B. Surface epithelium
 - C. Cortex
 - D. Medulla
 - E. Corticomedullary border
- (Periodic acid Schiff's, X400)



Figure 4: Photomicrograph of bursa of Fabricius in 4 week old Japanese quail

- A. Interfollicular septa
 - B. Surface epithelium
 - C. Cortex
 - D. Medulla
 - E. Corticomedullary border
- (Periodic acid Schiff's, X400)

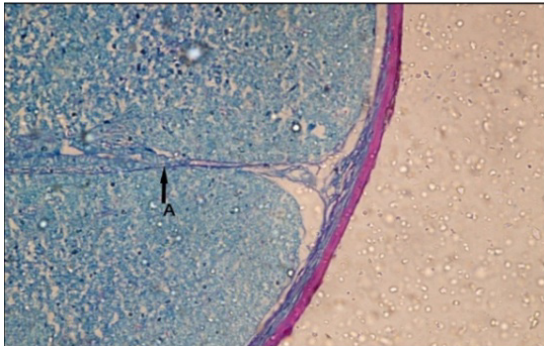


Figure 5: Photomicrograph of bursa of Fabricius in 1 week old Japanese quail

A. Interfollicular septa
(Alcian blue periodic acid Schiff's, X 400)

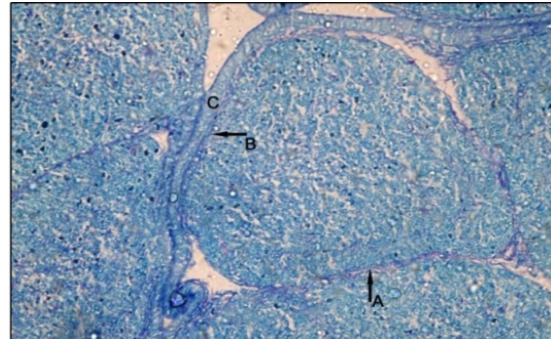


Figure 7: Photomicrograph of bursa of Fabricius in 3 week old Japanese quail

A. Interfollicular septa
B. Basement membrane
C. Surface epithelium
(Alcian blue periodic acid Schiff's, X 400)

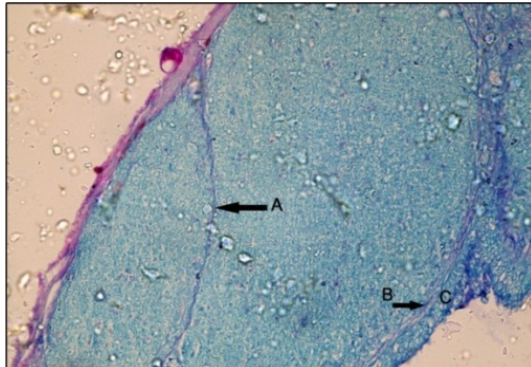


Figure 6: Photomicrograph of bursa of Fabricius in 2 week old Japanese quail

A. Interfollicular septa
B. Basement membrane
C. Surface epithelium
(Alcian blue periodic acid Schiff's, X 400)

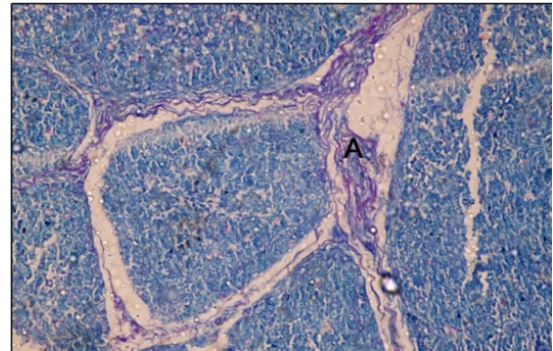


Figure 8: Photomicrograph of bursa of Fabricius in 4 week old Japanese quail

A. Interfollicular septa
(Alcian blue periodic acid Schiff's, X 400)

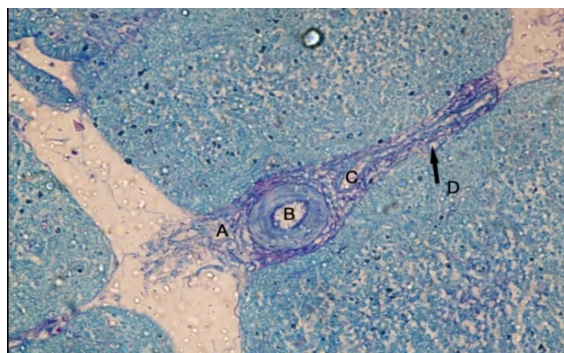


Figure 9: Photomicrograph of bursa of Fabricius in 2 week old Japanese quail

- A. Interfollicular connective tissue septa
 - B. Artery
 - C. Vein
 - D. Capillary
- (Alcian blue periodic acid Schiff's, X 400)

5, 6, 7 and 8). Similarly, high ABPAS activity was observed in the basement membrane of surface epithelium in group I birds which gradually decreased with the age. The apical border of the surface epithelium showed the high ABPAS reaction in all age groups except the group IV (Fig 6 and 7). The capillaries, tunica intima and adventitia of blood vessels in all age group of birds showed high to intense activity for acid mucopolysaccharides during the present study (Fig 9).

The observations recorded in the present study are in agreement with Jain *et al.* (2010) who reported the presence of ABPAS positive material in bursal epithelial surface in poultry birds. In line with present findings Singh *et al.* (2019) reported the gradual decrease in acid mucopolysaccharides activity

in interfollicular connective tissue with the advancement of age in bursa of Chabro bird. However, they recorded mild acid mucopolysaccharides activity in interfollicular connective tissue. This difference may attribute to the species differences.

More ABPAS activity in interfollicular connective tissue septa, epithelial basement membrane and apical border of epithelium may be correlated with the necessary physiological requirement during early stage of growth of organ.

CONCLUSION

The PAS activity varying from low to intense was present in almost all compartments of bursa. It might be suggestive for source of energy required for overall enhancement of metabolic process for tissue proliferation during bursal development. The presence of ABPAS activity in interfollicular connective tissue septa, epithelial basement membrane, and apical border of epithelium may be correlated with the necessary physiological requirement during early stage of growth of organ.

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