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Identification of histochemical constituents in bursa of Fabricius and thymus by staining methods in Broad Breasted Bronze turkeys

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Abstract

In bursa of Fabricius, histochemical staining revealed that the surface epithelium of bursal plica showed intense reaction for the presence of carbohydrate in upper half of surface epithelium whereas acid mucin was noticed only at apical part as a thin coating. The mucoid and epithelial cysts which appeared in the lymphoid follicles during involution showed intense reaction for the presence of carbohydrate. In thymus, Hassall's corpuscle showed intense reaction for carbohydrate and acid mucopolysaccharides. The plasma cells showed strong PAS positive Russell granules in their cytoplasm. In both the organs, the positive reaction for lipids were observed during involution.

Keywords: Bursa of fabricius, Thymus, Turkey, histochemistry

Introduction

In poultry industry, the turkey is one of the diversified species plays a substantial role in economic status and viable livelihood for small and marginal farmers (Prabakaran *et al.*, 2020) [2]. Although turkeys are resistant to most of the diseases, they are affected by few economically important diseases. Hence, greater emphasis should be given to prevention which needs thorough understanding of immune organs. The primary immune organs such as bursa of Fabricius and thymus plays a pivotal role in the maintenance of optimum health by protecting from diseases and adverse abiotic factors (Penchev, 2020) [3]. The histoarchitecture of spleen and harderian gland are constituted by cells, stromal fibres and histochemical components. Understanding the histochemical reactions of these organs to various stains helps to analyze its proportions in these organs and resolve its complete microanatomy. With this scope, this study was formulated to reveal the histochemistry of these immunological organs in turkey.

Materials and Methods

The bursa of Fabricius and thymus of turkey were collected from adult Broad Breasted Bronze variety of turkeys. The organs were fixed in various fixatives such as 10 per cent Neutral Buffered Formalin (NBF) to demonstrate the histochemical components by various staining methods. The paraffin sections of 3-5 µm thickness were made using a Leica microtome (RM-2145) for histochemical staining methods and 15-20 µm thickness frozen sections made by cryostat were used to demonstrate the lipids in immune organs. The following staining techniques were employed.

- Periodic acid-Schiff technique for carbohydrates (Bancroft and Stevens, 1996) [4].
- Alcian blue technique pH 2.5 for acid mucins (Bancroft and Stevens, 1996) [4].
- Combined Alcian blue-PAS technique for acid and neutral mucins (Bancroft and Stevens, 1996) [4].
- Oil red O method for lipids (Bancroft and Stevens, 1996) [4].

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Results and Discussion

Bursa of Fabricius

Histochemical staining revealed that the surface epithelium of bursal plica showed intense reaction for the presence of carbohydrate in upper half of surface epithelium whereas acid mucin was noticed only at apical part as a thin coating (Fig 1). In concurrence, the surface epithelium of bursal plica showed positive reaction for PAS and alcian blue in domestic fowl (Leena *et al.*, 2009) ^[5]. But, Jain *et al.* (2010) ^[7] demonstrated mild to moderate PAS and combined AB-PAS activity in the epithelium of bursa in CARI Shyama and Vanaraja breeds. Karadag *et al.* (2015) ^[1] found weak PAS reaction in the follicle associated epithelium of bursa of Fabricius in long legged buzzard.

The mucoid cysts which appeared in the lymphoid follicles during involution showed intense reaction for the presence of carbohydrate (Fig 2). In contrary, mucoid substance appeared during involution revealed positive reaction for acid mucopolysaccharides in domestic fowl (Leena *et al.*, 2009) ^[5]. Moderate reaction was noticed in the connective tissue at interfollicular septa, sub-epithelial region and cortico-medullary border. The reticular cells and reticular structures showed mild PAS positive reaction. Leena *et al.* (2009) ^[5] noticed PAS reaction in the cortico-medullary border in domestic fowl. In accordance to the present study, Mahanta *et al.* (2020) ^[8] in local hill fowl and Rhode Island Red reported moderate PAS activity in plical connective tissue at center and trabeculae. As observed by Karadag *et al.* (2015) ^[1] in long legged buzzard, the smooth muscle at the wall of bursa of Fabricius showed weak PAS positive activity.

The epithelial cysts formed at involutory phase indicated intense reaction for carbohydrate (Fig 3). Both acid and neutral mucopolysaccharides were noticed in the epithelial cysts as demonstrated by Mahanta *et al.* (2020) ^[8] in local hill fowl and Rhode Island Red. But, Leena *et al.* (2009) ^[5] observed strong reaction in infolded epithelium for acid mucopolysaccharides in domestic fowl. Also in contrast, Leena *et al.* (2012) ^[6] in domestic fowl of bursa found alcian blue positive material in the simple tubular gland lined by columnar cells formed during 10th week of age.

The parenchyma of bursa of Fabricius at ten months of age showed large adipose tissue accumulation within the fibrous proliferation. In accordance, localized lipid deposition was noticed in the lamina propria in 10th and 18th week and more adipose cells were noticed from 22nd-24th week of age in domestic fowl (Leena *et al.*, 2009) ^[5].

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Thymus

The PAS reaction for carbohydrate showed moderate activity in stromal components as observed by Mahanta *et al.* (2020) ^[8] in local hill fowl and Rhode Island Red and mild activity in reticular structures and Hassall's corpuscles type I, III and IV. The type II Hassall's corpuscle showed intense reaction for carbohydrate and acid mucopolysaccharides. The neutral mucopolysaccharides activity was intense in type V and type VI Hassall's corpuscle (Fig 1, Fig 2). Whereas, Gulmez and Aslan, (1999) ^[9] in native geese, Leena *et al.* (2009) ^[5] in domestic fowl and Mahanta *et al.* (2020) ^[8] in local hill fowl and Rhode Island Red demonstrated moderate to intense PAS and alcian blue reaction in Hassall's corpuscles, but not mentioned the types.

The plasma cells showed strong PAS positive Russell granules in their cytoplasm as mentioned by Bejdic *et al.* (2014) ^[10] in Harderian gland of laying hens (Fig 3). Moderate PAS reaction was noticed in the heterophils at reticular structures of thymus. Cunhal *et al.* (2021) ^[11] stated that the heterophils were PAS positive. Also he explained that the presence of glycogen in heterophils might indicate the phagocytosis mechanism is energy dependent and the intensity of reaction to PAS increases with cell maturation due to accumulation of the polysaccharide.

The capsule and interlobular septa of thymus at terminal stage of involution showed more fatty infiltration. It concurs with Leena *et al.* (2009) ^[5] in domestic fowl who observed faint diffuse lipid droplets, localized and widespread accumulation of adipocytes at septa in thymus during involutory stage by lipid histochemistry.

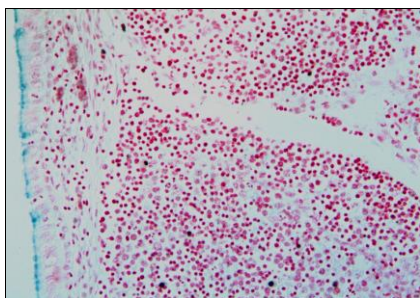


Fig 1: Strong reaction for acid mucopolysaccharides in the surface epithelium of bursa of Fabricius in turkey Alcian blue x 400

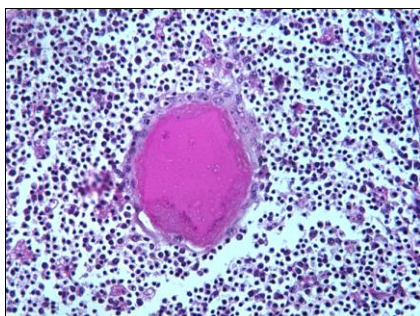


Fig 2: Strong reaction for neutral mucopolysaccharides in the mucoid cysts of bursa of Fabricius in turkey PAS x 400

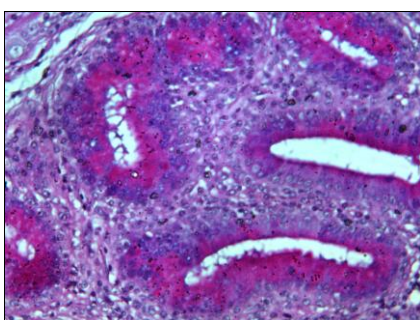


Fig 3: Strong reaction for neutral mucopolysaccharides in the epithelial cysts of bursa of Fabricius in turkey PAS x 1000

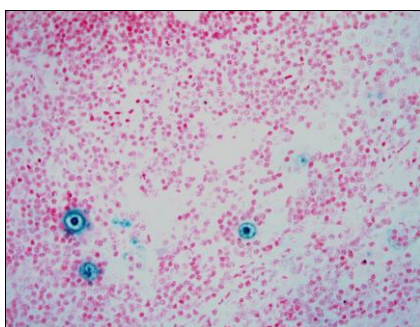


Fig 4: Strong reaction for acid mucopolysaccharides in the Hassall's corpuscles in thymus of turkey Alcian blue x 400

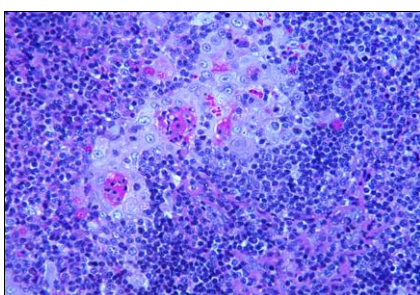


Fig 5: Strong reaction for neutral mucopolysaccharides in the Hassall's corpuscles in thymus of turkey PAS x 1000

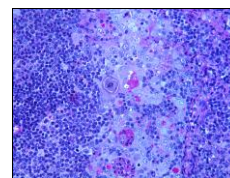


Fig 6: Strong PAS reaction for neutral mucopolysaccharides in the plasma cells of thymus in turkey PAS x 1000

Conflict of Interest

Not available

Financial Support

Not available

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