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# HISTOMORPHOLOGICAL CHARACTERIZATION OF THE THYMUS GLAND IN KADAKNATH BIRDS

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# ABSTRACT

The histomorphological study of thymus was carried out in Kadaknath birds of different ages, ranging from day old to twenty six week old irrespective of sex. After collection of specimen, the tissue samples were fixed in neutral buffered formalin. The fixed samples were processed for routine tissue preparation technique. After routine technique, the tissue samples were stained by different method. The thymus gland was encapsulated by thin connective tissue capsule along with adipose tissue. Few Hassall's corpuscles were appeared at second week of age. Two types of Hassall's corpuscles were seen as type-I and type-II. Three types of myoid cells were found as type-I, type-II and type-III. Presence of melanocytes in thymus was unique characteristic feature of present study, this was fusiform cell with elongated or round nuclei occurred in cytoplasm. The melanoytes generally occurred in all component of thymus. There was no regression of thymus after 26<sup>th</sup> week of age.

KEYWORDS: Kadaknath, thymus, histology.

# INTRODUCTION

The Kadaknath is mostly found in Madhya Pradesh especially around Jhabua and Dhar district (Pandey et al., 2002). The lymphoid system of birds is consisting of unique organs and divided into two morphologically and functionally distinct components. The thymus and bursa of chicken is considered to be a 'central or primary' lymphoid tissue, whereas spleen, cecal tonsil and mucosaassociated lymphatic tissues (MALT) are considered as 'peripheral or secondary' lymphoid tissues. The thymus, which produces T lymphocytes and responsible for cellular immunity, in contrast, bursa of Fabricius responsible for the production of B lymphocytes causes humoral immunity. Thymic lobes are located parallel to the vagus nerve and internal jugular veins (Khan et al., 2014). On each side of the neck there are 6-8 separate lobes, extending from the third cervical vertebra to the upper thoracic segments (Akter et al., 2006).

#### **MATERIALS & METHODS**

The study was done on thirty samples of thymic tissue collected from five different age groups ranging from day old to twenty six weeks old male and female birds of Kadaknath breed of poultry. Six birds were used in each age group. These birds were sacrificed ethically and for histology, tissue pieces were fixed in 10% neutral buffered formalin for 24 hours. Fixed thymus tissues were processed by routine paraffin embedding technique and paraffin sections of 5 to  $7\mu$  were taken. The paraffin sections were stain by different histological method as Hematoxyline & Eosin for normal histological structure, Verhoeff's method for elastic fibers, Van-Gieson for collagen fibers (Singh and Sulochana, 1997) and Gomori's method for reticular fibers (Drury and Wallington, 1980).

# **RESULTS AND DISCUSSION**

The thymus of Kadaknath was encapsulated by thin connective tissue capsule along with adipose tissue (fig. 1). From connective tissue, septae arised entered into lobes dividing them into lobules, along with septae, blood vessels and nerve also entered into lobules. These observations are similar to those reported by (Senapati *et al.*, 2015) in quail, chicken and duck. The connective tissue capsule specially made up of thin irregular bundles of collagen fibers along with few elastic and reticular fibers which was similar to the finding (Kannan *et al.*, 2017) in layer chicken.

At the age of one week of Kadaknath birds, the thymus showed more lobulation with outer dark cortex, filled with lymphocytes and inner light medulla formed of large lymphocytes with centrally placed nuclei and acidophilic cytoplasm. These observations are similar to those reported by Treesh *et al.* (2014) in chicken. Division of the cortex and medulla started at first week and was distinct at fourth week of age. These finding differ from Giriraja birds (Chandrashekhar *et al.*, 2008) who reported that division of the cortex and medulla started at first week and was distinct at third week of age.

During the first week the cortex contained huge pale eosinophilic structures, containing more than one thymocytes were frequently undergoing mitotic divisions. These finding are in agreement with Chandrashekhar *et al.* (2008) in Giriraja birds. It is due to the fact that cortical tissue contained immature lymphocytes *i.e.* thymocytes undergoes development.

Few Hassall's corpuscles in medulla and blood capillaries at the corticomedullary junction were appeared at second week of age. These findings are differ from Treesh *et al.* (2014) in chicken who founded that the Hassall's corpuscles and blood capillary were appeared at four week of age, Islam *et al.* (2011) in native chicken who founded that medulla was pale stained with few Hassall's corpuscles at embryonic day (ED20). (Kannan *et al.*, 2015) in Nandanam chicken revealed that presence of Hassall's corpuscles was also noticed in the medulla as well as cortical areas of day-old, four weeks and ten weeks age groups. The two type of cyst as intracellular and intercellular cyst were associated with Hassall's



**FIGURE 1**: Photomicrograph of thymus (3<sup>rd</sup> week) showing capsule (C), Cortex, Medulla and septae ( H & E 407 x)

In the present study, two types of Hassall's corpuscles were seen as type-I flattened reticuloepithelial cells, which were arranged in concentric manner around the central cornified mass which was acidophilic in nature (Fig. 2) whereas type-II Hassall's corpuscles were spherical in shape encircled by flattened reticuloepithelial cells, enclosing a hyaline mass with a group of cells (Fig. 3). These results differ from Muthukumaran et al. (2011) in turkey who observed three types of Hassall's corpuscles as Hassall`s corpuscles contained type-1 flattened reticuloepithelial cells were arranged in concentric manner around the central keratinized mass, type-2 Hassall's corpuscles were spherical in shape lined by flattened reticuloepithelial, enclosing a hyaline mass with a group of



**FIGURE 3**: Photomicrograph of thymus showing type II Hassall's corpuscles in Medulla ( H & E 804 x)

In fourth week of age (group-II), many lobules possessed a common medulla and by the end of the  $8^{th}$  week, the

corpuscles similar observation was reported Kannan *et al.* (2015) in Nandanam chicken. The mitotic index of the cortex was high in the first two groups then it decreased, however at fourth group get increased mitotic index which may be due to the effect of prolactin. These finding were consistent with Chandrashekhar *et al.* (2008) in Giriraja birds.



**FIGURE 2**: Photomicrograph of thymus showing type I Hassall's corpuscles in Medulla ( H & E 804 x)

cells and type-3 Hassall's corpuscles were composed of flattened reticuloepithelial, enclosing a hyaline mass alone.

The skeletal myofibril cells *i.e.* myoid cell were commonly occurred in thymic medulla. In the present study, minimum three types of skeletal muscle cells were found as type-I cells was round and ovoid in shape and striated myofibrils encircled the nucleus, type- II cells were elongated and their end was appeared as Y shaped and type -III was triangular in shape with the round and concentrally placed nucleus (Fig. 4). However Davison *et al.* (2008) reported two types of myoid cells in avian thymus. The roles of these myoid cells were not clearly identified.



**FIGURE 4**: Photomicrograph of thymus (15<sup>th</sup> week) showing type I, type II and type III myoid cell capsule (C), Cortex, Medulla and septae ( H & E 1615 x)

medulla had complete division by interlobular connective tissue. These finding differ from Chandrashekhar *et al.* 

(2008) in Giriraja birds who reported that at the third week of age many lobules possessed a common medulla and by the end of the sixth week, the medulla had division by interlobular connective tissue. These finding in present study showing initiation of division of medulla by interlobular connective tissue started at similar age but it were lasting higher age of Kadaknath birds.

The involuntary changes were seen from eight week onwards which progressed till 16<sup>th</sup> week of age, after which enlargment and alteration of thymic architecture was noticed. The involuntary changes in the thymus gland was characterized by loss of interlobulation septa, pyknosis of thymocytes, extension of area of medulla, increase in size and number of Hassall's corpuscles and myoid cells (fig. 5). These finding differ from Chandrashekhar *et al.* (2008) in Giriraja birds who reported that the involuntary changes were seen from eight week onwards which progressed till  $14^{th}$  week of age, after which enlargment and resortion of thymic architecture was noticed. The diameter of lymphocytes in cortex and medulla were observed insignificantly. The numbers of Hassall's corpuscles increased with advancement of age similar observation was accordance with Kannan *et al.* (2015) in chicken. The number of corpuscle was found to be more in advanced age which indicated the involuntary changes in aged birds.



**FIGURE 5**: Photomicrograph of thymus (16<sup>th</sup> week) showing involuntary changes and melanocytes ( M) type I, type II and type III myoid cell capsule (C), Cortex, Medulla and septae ( H & E 1615 x)

Presence of melanocytes in thymus was unique characteristic feature of present study; this was fusiform cell with elongated or round nuclei. The melanoytes generally occurred in thin connective tissue capsule and septae. The melanocytes were higher in group-IV (fig. 5). These finding were not available in other literature. Due to melanocytes deposition in organs the external extremities as well as internal organs is black in color. The thymus contained immature erythrocytes. These finding might be link to erythropoeitic function and closely associated with avian breeding cycle, similar fact noted by Ciriaco et al. (2003) in birds. No regression was observed at  $26^{th}$  week of age. This observation was differ from Khenenou et al. (2012) in broiler chicken who reported that regression of thymic lobules started at fourteen week of age due to invasion of the parenchyma by adipose tissue. At the 23rd week, a very significant interstitial adipose tissue and degeneration of the lobules was noticed. These finding may be due to well knowing fact about disease resistance of Kadaknath birds is well disease resistance. The numbers of Hassall's corpuscles increased with advancement of age (Fig. 5,) similar observation was accordance with Kannan et al. (2015) in chicken. The number of corpuscles was found to be more in advanced age which indicated the involuntary changes in aged birds.

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