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Gross and Microanatomical Study of the Cornea in Japanese quail (*Coturnix coturnix japonica*)

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A B S T R A C T

The present study was carried out on cornea of the Japanese quail. The cornea was the transparent, non-vascular structure and slightly curved front part of the eye and it covers the anterior chamber, iris and pupil in back side. The cornea was composed of anterior epithelium, bowman's membrane, substantia propria, descemet's membrane and posterior epithelium. The anterior epithelium was lined by non-keratinized stratified squamous epithelium of five to six layers of epithelial cells. The bowman's membrane was observed as an acellular, transparent homogenous layer with numerous collagen fibres, which ran parallel to the corneal surface and placed beneath the anterior epithelium. The thickness of the substantia propria was found to be more than the other layers of the cornea and hence formed the greater part of the cornea. The posterior epithelium which lined the inner side of the descemet's membrane consisted of a single layer of cuboidal cells. The thickness of the cornea ranged from 110.20±0.209µm to 156.39 ±1.244µm on the right eye ball and 109.70±0.182µm to 156.26±1.199µm on the left eye ball in two week-old to fourteen week-old Japanese quail.

Introduction

Quail is one of the smallest avian species of the pheasant family. The quail has been first domesticated in Japan in 1595. In India the bird has been domesticated in 1974. Two species of quail are found in India one is black breasted quail found in jungle (*Coturnix coromandelica*) and the other species is the brown colour Japanese quail (*Coturnix coturnix japonica*) which is bred for meat for commercial production (Priti and Satish, 2014). The eye is one of the most complex organs in the body which gives us a sense of sight allowing birds, animals and humans to learn more about the surrounding world. Vision is the major sense by which birds perceive their environment (Olopade *et al.,* 2005 and Hall and Ross, 2007).

While the cornea contributes most of the eye's focusing power, its focus is fixed. The cornea is avascular, thin and more strongly curved transparent membrane, which acts as the major structure of reflection in the eye of birds. Although the structure of the cornea has been studied, the literature on the age related on the histology and histometry is very scanty in Japanese quail. Hence, the present work was undertaken with the objective of studying the detailed gross morphology, histology, histometry and functional aspect of the cornea in Japanese quail for better understanding of the structure of eye ball and vision in Japanese quail.

Materials and Methods

Ethical approval for the study was obtained from the ethical committee of the madras veterinary college, TANUVAS, Chennai-7. The materials for the present study, the Japanese quail were procured from Poultry research station, Madhavaram milk colony, Chennai-51. The materials for the research work were collected from six birds each from viz., Two weeks, Eight weeks and fourteen weeks of age. Each group consisted of three male and three female birds. Both right and left eye ball were collected and their gross morphometrical measurements were recorded for all the birds. The eye balls were fixed in the Modified Davidson fixative and the tissue samples were processed as per the standard procedures (Spencer and Bancroft, 2013). Sections of 5-6 µm thickness were used for the routine and special staining techniques. The thickness of the cornea of the eyeball were measured and recorded by using the oculometer.

Results and Discussion

Gross morphology

In the present study, the cornea was transparent (Fig. 1), non-vascular structure and slightly curved in all the age groups studied and these findings are similar to that of Maggs *et al.,* (2013) in birds. Cornea acts as barrier to the external environment serves as a major refractive component of the eye of the chicken (Ritchy *et al.,* 2011). The cornea acts as the eye’s outermost lens. It functions like a window that controls and focuses the entry of light into the eye. The cornea contributes between 65-75 per cent of the eyes total focusing power (http://www.nei.nih.gov/ health/corneal disease).

The corneal diameter of the both eyes was observed to increase from two week to fourteen week-old Japanese quail. However, no significant difference was noticed between members of same age group in the corneal diameter of right and left eyes (Table 1). An enlarged cornea is said to allow more amount of light to enter the eye through an enlarged pupil. The corneal diameter can be associated with visual sensitivity, the light-gathering ability of the eye (Hall and Ross, 2007).

Histology

In the present study, the cornea of the Japanese quail was composed of five layers *viz.,* the anterior epithelium, the Bowman’s membrane, the substantia propria, the Descemet’s membrane and the posterior epithelium in all the age groups (Fig. 2). The present observations are in total agreement with the findings of the Goncalves *et al.,* (2016) in Japanese quail.

The anterior epithelium was lined by non-keratinized stratified squamous epithelium of five to six layers of cells was observed in all the age groups studied. The basal cell layer was made up of tall columnar cells with oval nuclei towards the apex of the cell. Small polygonal cells with oval nuclei interrupted the basal cells.

The stratum basale was overlaid by three to four layers of flattened cells (Fig. 3). These findings are in harmony with the observations of Hodges (1974) and Goncalves *et al.*, (2016) in birds.

Bowman's membrane was observed as an acellular, transparent homogenous layer with numerous collagen fibres, which ran parallel to the corneal surface and placed beneath the anterior epithelium (Fig. 2). This observation is similar to the findings of Goncalves *et al.*, (2016) in Japanese quail and contrary to the findings of Fitzgerald (1969), who as stated that the Bowman's membrane is absent in quail. The Bowman's membrane act as an important protective layer of the cornea to prevent the bacterial invasion as per the statement of Wilson and Hong (2000) in chicken.

Table.1 Corneal diameter of right and left eye balls of Japanese quail in different age groups (Mean ± SE)

SI. No	Age groups	Number	Right Corneal Diameter (mm)	Left Corneal Diameter (mm)
1.	2 week	6	$4.86^{a} \pm 0.111$	$4.68^{a} \pm 0.098$
2.	8 week	6	$6.41^{b} \pm 0.113$	$6.23^{b} \pm 0.084$
3.	14 week	6	$6.98^{c} \pm 0.119$	$6.88^{c} \pm 0.119$
F-Value			90.805**	123.629**

Mean bearing same superscript did not differ significantly
** - Highly significant (P≤0.01)

Table.2 Thickness of cornea in right and left eye balls of Japanese quail in different age groups (Mean ± SE)

SI. No	Age groups	Number	Right Cornea (µm)	Left Cornea (µm)
1.	2 week	6	$110.20^{a} \pm 0.209$	$109.70^{a} \pm 0.182$
2.	8 week	6	$149.86^{b} \pm 0.671$	$149.48^{b} \pm 0.641$
3.	14 week	6	$156.39^{c} \pm 1.244$	$156.26^{c} \pm 1.199$
F-Value			917.24**	1008.39**

Mean bearing same superscript did not differ significantly
** - Highly significant (P≤0.01)

Fig.1 Photograph showing cornea (Arrow) of an eight week-old male Japanese quail

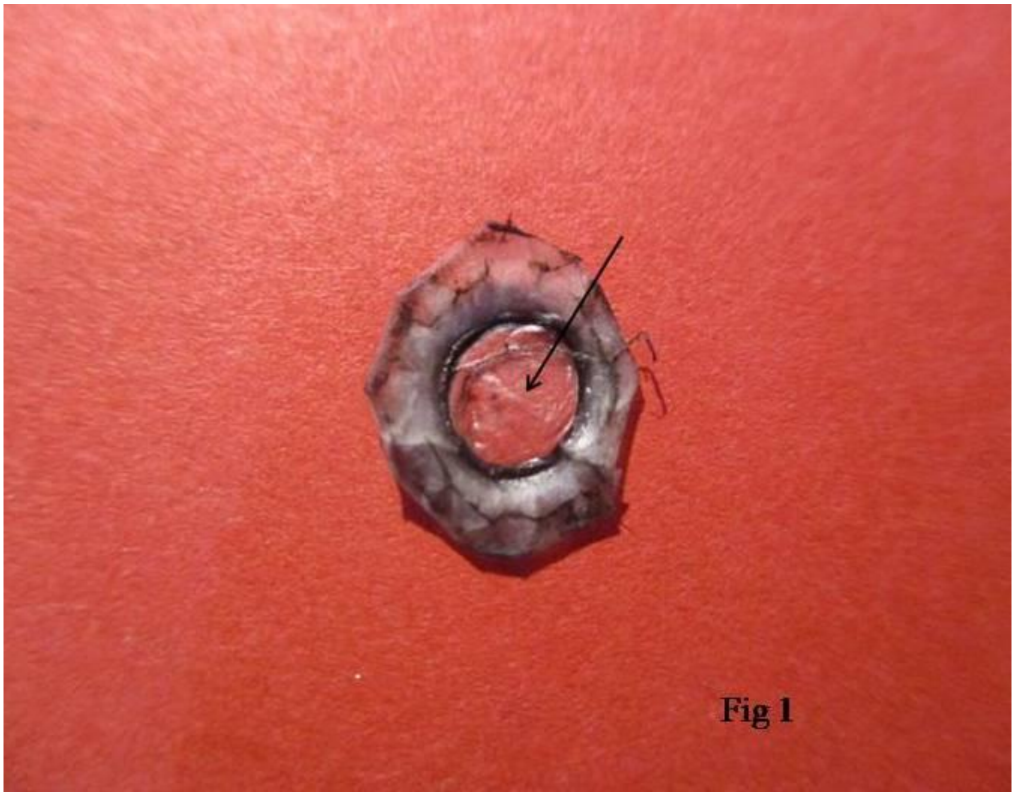

Fig.2 Photomicrograph of the cornea of the right eye ball in a fourteen week old male Japanese quail. A-Anterior Epithelium; B-Bowman's membrane; C-Substantia Propria; D-Descemet's membrane; E-Posterior Epithelium; I-Intercellular Space. H&E × 100

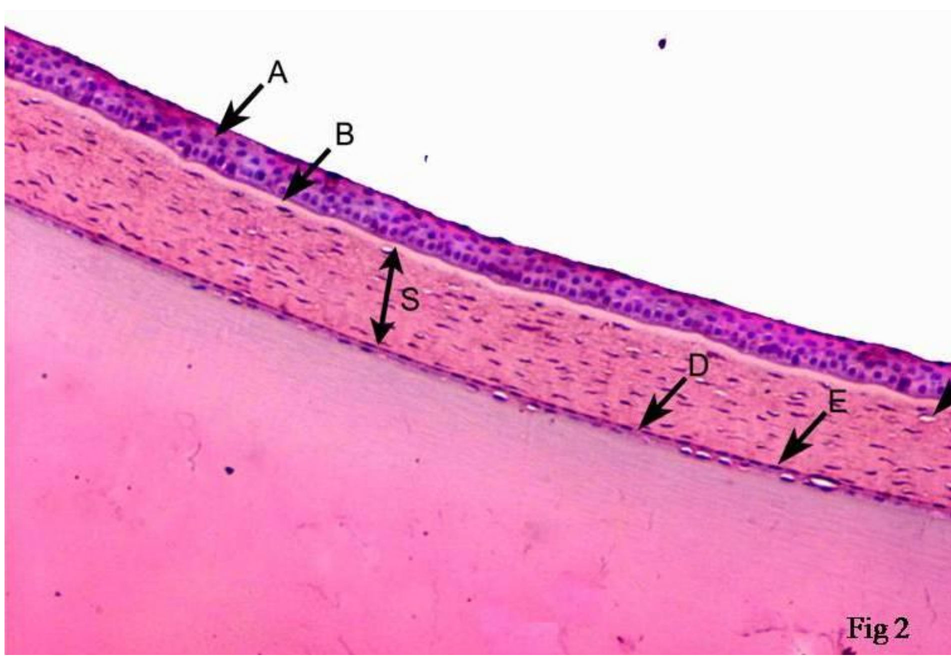

Fig.3 Photomicrograph of the cornea of the left eye ball in a eight week-old female Japanese quail. A-Anterior epithelium; S-Substantia Propria; TC-Tall columnar cell; FC-Flattened cell. H&E × 400

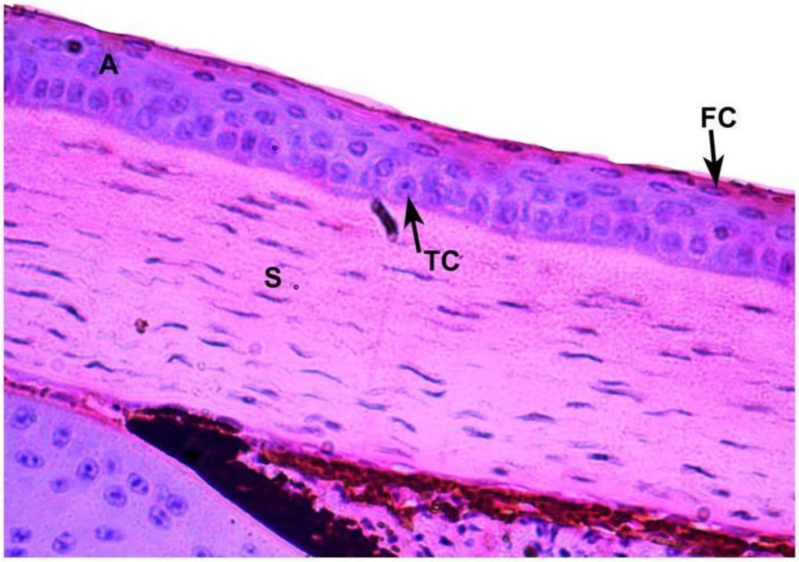

Fig. 3

Fig.4 Photomicrograph of the corneoscleral junction and associated structures of the left eye ball in a fourteen week-old female Japanese quail. Cf – Ciliary folds; Cr – Crampton's muscle; Br – Brucke's muscle; CS – Canal of Schlemm; CSJ – Corneoscleral junction; SF – Spaces of Fontana. H&E × 100

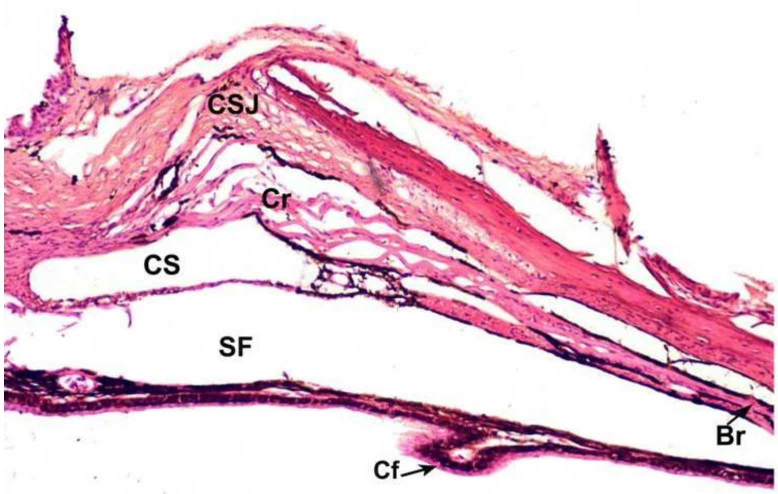

Fig 4

The substantia propria was transparent and not completely homogenous, as it was composed of bundles of collagen fibres with flattened fibroblasts observed parallel to the corneal surface. The thickness of the substantia propria was found to be more than the other layers of the cornea and hence formed the greater part of the cornea.

Numerous intercellular spaces were also seen in this layer (Fig. 2 and 3). This is in agreement to the finding of Fitzgerald (1969) in Japanese quail. The fibroblasts in the substantia propria are said to transport the nutrients and waste from the blood vessels in the Corneoscleral junction as per the statement of Glasser and Howland (1996) in domestic fowl. Further, the stromal collagen matrix provides mechanical strength to the cornea in owl (Jezler *et al.,* 2010).

Descemet's membrane was observed as a thin homogenous structure and placed beneath the substantia propria. The posterior epithelium which lined the inner side of the Descemet's membrane consisted of a single layer of cuboidal cells (Fig. 2) as per the finding of Goncalves *et al.,* (2016) in Japanese quail.

The thickness of the cornea ranged from 110.20±0.209µm to 156.39 ±1.244µm on the right eye ball and 109.70±0.182µm to 156.26±1.199µm on the left eye ball in two week-old to fourteen week-old Japanese quail (Table 2). Further, in the present study, the cornea was observed to be thinner at the middle and thicker at Corneoscleral junction in all the age groups.

In the present study, the Corneoscleral junction was composed of bundles of collagen fibres, elastic fibres and fibroblasts. A loose network of fine elastic fibres covered by the endothelial cells, the pectinate ligament passed across from the ciliary body and the base of the iris to the base of the cornea. In all the age groups, numerous discrete endothelial lined spaces, the spaces of Fontana (Fig. 4) were also observed between the fine elastic fibrils of the pectinate ligament.

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