Iraqi Journal of Veterinary Sciences

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Anatomical and histological structure of the cornea in Sparrow hawk *Accipiter nisus*

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Article information

Article history:
Received April 19, 2020
Accepted May 11, 2020
Available online June 23, 2021

Keywords:
Cornea
Bowman's membrane
Descemet membrane
Sparrow hawk

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Abstract

The current study conducted to investigate the anatomical and histological structure of the cornea in sparrow hawk *accipiter nisus*. By using an optical microscope and histological stains. Anatomical results showed that the cornea composed of two parts, the peripheral cornea and the central cornea, and there was variation in corneal thickness according to the anatomical region. Histologically, the cornea consists of five layers. The first layer of the central cornea consists of non- keratinized stratified squamous epithelial tissue composed of four rows of cells. Whereas, Bowman's membrane appeared clear and thicker on the dorsal side than on the ventral side. The corneal stroma consists of collagen fibers arranged in three secondary layers within the stroma. The corneal stroma also distinguished by the presence of fibroblast cells, which randomly distributed among the collagen fibers. The Descemet membrane appeared clear and equal in thickness between the dorsal and ventral sides. The endothelium tissue appears in the form simple squamous epithelia. The histological structure of the cornea root seemed similar to its formation in the central cornea. Still, it characterized by the presence of pigment cells spread in the stromal corneal root. Among the distinct results that appeared in this study is the presence of blood vessels spreading in the root of the cornea. The study concluded that the anatomical and histological structure of the cornea in this bird distinguished by some specialties that contribute to increasing the effectiveness of the function of the cornea in this bird.

DOI: 10.33899/ijvs.2020.126976.1424, ©2021, College of Veterinary Medicine, University of Mosul.

Introduction

The visual requirements in vertebrates differ significantly, and this is evident through the development of their optical parts in response to these requirements and in enhancing the performance of their visual behavior (1). Birds are among the most vertebrates dependent on the sense of sight, and this evidenced by the large size of their eyes and their visual system very different from other vertebrates (2). Birds have eyes that can distinguish distances well so as not to collide with trees and other objects, and this is evident in the center of sight, which makes up half of the brain of birds. Also, each bird has its unique fingerprint, it can never be the same in another bird because it is like the fingerprint of the hand, which can never be similar (3). The sense of sight in birds, especially wild ones, and types of birds of prey are among the most important major senses. The large eye size in birds compared to the size of the body is a good indicator for understanding this importance. It is necessary to have a sound, good, and undamaged sense of vision to develop natural behavior in birds of prey, such as independent feeding and aviation, as the total or partial damage to the eyesight in birds greatly affects their ability to direct themselves in the atmosphere, respond to external stimuli or Environmental changes especially while hunting (4). All birds have similar basic makeup eyes, but many differences reflect their environmental needs (5). The eyeball in birds, as in other vertebrates, is anatomically and histologically consist of three layers (Tunicas). The first layer is the tunica fibrosa (6). The second layer is Tunica vasculosa, while the third layer is Tunica Nervosa. The cornea is the anterior, outer portion of the fibrous tunica of the vertebrate's eye. It characterized by being avascular and transparent, and it depends on the aqueous fluid and blood vessels surrounding its ends to provide it with nutrients. In swimming birds, the cornea is small, as it forms a relatively small area of the eye. The cornea is thin and arched, but it is convex and actively in many birds such as Strigiform. In general, the cornea is more transparent in birds than in other vertebrate species (7). Jezler *et al.,* (8) mentioned that the cornea in the eagle and the owl is transparent and strongly curved, and this allows a sufficient amount of light to be refracted in it and sufficiently to form an image on the retina. The Eurasian sparrow hawk *Accipiter nisus* described scientifically for the first time during the eighteenth century by the Swedish scientist Carlos Linnaeus. The Eurasian sparrow hawk called in the city of Mosul/ Iraq sparrow hawk. This bird belongs to the order of Accipitriformes, family Accipitridae. Eurasian sparrow hawk is a widespread species across most of the temperate and subtropical regions of the ancient world and is one of the most common prey in Europe (9).

Therefore, the objective of this study was to identify the anatomical and histological structure of the cornea in sparrow hawk using many histological stains.

Material and methods

Ten healthy and adult of *A. nisus nisus* obtained (five from each gender), these birds were carried to the laboratory of anatomy and histology / Biology Department / Education College for Pure Sciences, Mosul University. They were placed in a metal cage of appropriate size for three days before dissection to ensure their safety, the birds fed on small animals such as finches and small pieces of meat (10). The birds anaesthetized with Chloroform, then dissected. After that, the head cut, then the feathers removed, the orbit' bones were cut, then finally the eyeball was removed from the Orbit quarry using fine tweezers (11).

The eyes washed with a physiological solution and then transferred to a petri dish containing a physiological solution. The wax embedding method used to prepare tissue sections in this study, and the microscopic slides were made based on the technique of (12). The histological sections stained with the following stains: Delafield's Hematoxylin and Eosin stain (H&E), Mallory's Trichrome stain (TS), Periodic Acid - Schiff technique (PAS) and Toluidine Blue (TB) (12-15). The micromorphometric was taken based on the method of (16).

Results

The cornea appeared in a convex outward and anatomically composed of two parts: the peripheral cornea or the root of the cornea, which represents the part of the cornea that connected to the sclera. The second part is known as the central cornea and is opposite to the iris and the pupil. The cornea root appeared less thick than the central cornea, and its average thickness in both sides was 282.870 ± 0.32 µm, while the thickness of the central cornea in the dorsal side was 593.185 ± 0.54 µm, and the average thickness of the central cornea in the ventral side was 577.074±0.43 µm (Figures 1-4).

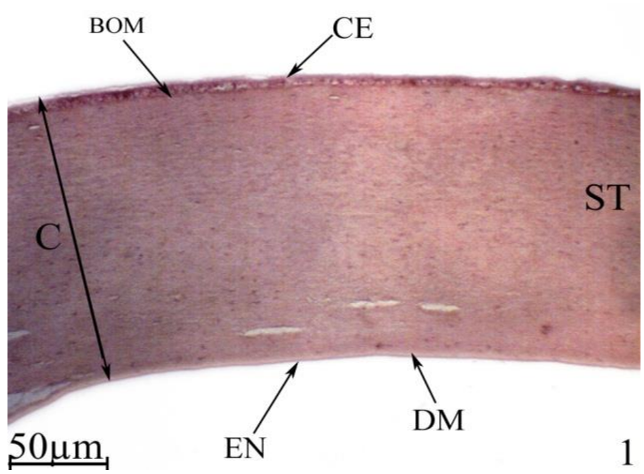

Figure 1: Cross-section in the central cornea (C) in dorsal side. (CE) Anterior Corneal epithelium. (BOM) Bowman's membrane. (ST) Stroma. (EN) Endothelium. (DM) Descemet membrane. PAS stain.

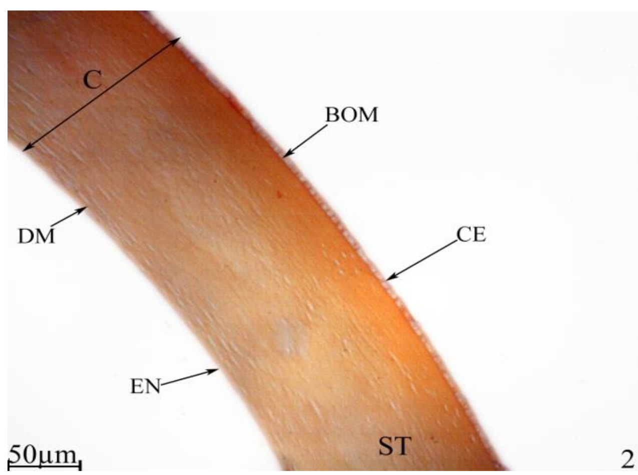

Figure 2: Cross-section in the central cornea (C) in dorsal side. (CE) Anterior Corneal epithelium. (BOM) Bowman's membrane. (ST) Stroma. (EN) Endothelium. (DM) Descemet membrane. TS stain.

Histologically, the central cornea composed of a layer of epithelial tissue based on the basal membrane. Bowman's membrane followed the epithelial tissue, which in turn followed by an internal stroma that consists of several layers of collagen fibers. The stroma, in turn, based on a membrane called a Descemet membrane that lines the endothelium (Figures 1-3). While the cornea root appeared similar to the central cornea histologically, it consists of the layers mentioned above itself as well as it contains pigment cells scattered and in the form of alternate rows and randomly distributed in the stroma. The collagen fibers of the corneal stromal are loose in the middle and compact at both ends of the cornea root. One of the significant results that appeared in the cornea root of this bird is the presence of blood vessels distributed geometrically in this region. These vessels are relatively large, and the diameter of larger one was 48.472 ± 0.51 µm (Figures 5 and 6).

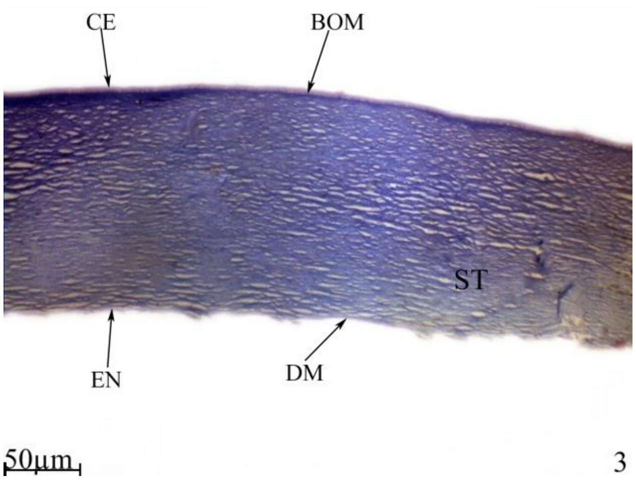

Figure 3: Cross-section in the central cornea in ventral side. (CE) Anterior Corneal epithelium. (BOM) Bowman's membrane. (ST) Stroma. (EN) Endothelium. (DM) Descemet membrane. TB stain.

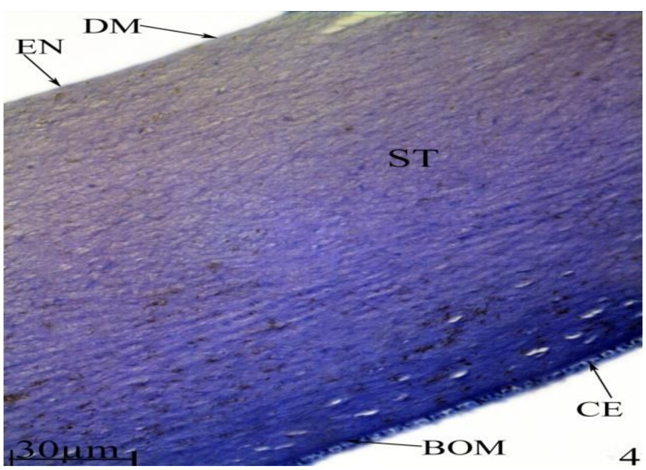

Figure 4: Cross-section in the central cornea in ventral side. (CE) Anterior Corneal epithelium. (BOM) Bowman's membrane. (ST) Stroma. (EN) Endothelium. (DM) Descemet membrane. TB stain.

The epithelial tissue of the central cornea was formed of the non-keratinized stratified squamous type, which consists of 4 layers of cells. The basal cells are square to vertical, with an average length is 11.341± 0.91 µm, while the average width is 9.879 ± 0.341 µm. The nuclei of this cell are spherical and have an average diameter of 4.756 ± 0.21 µm. The cell cytoplasm is clear transparent. The layer of the basal cells follows a layer of small polygonal cells whose dimensions cannot be recognized, followed by a layer of squamous cells and then non-keratinized squamous cells (Figures 7 and 8). These cells based on a straight, non-wavy basal membrane with a thickness of 2.315 ± 0.046 µm.

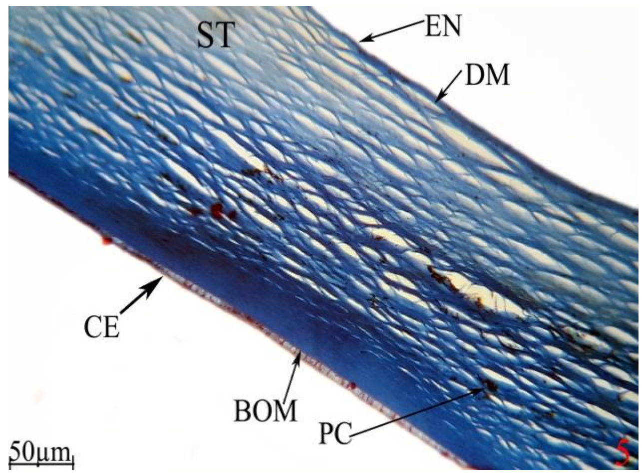

Figure 5: Cross-section in the Root of cornea. (CE) Anterior Corneal epithelium. (BOM) Bowman's membrane. (ST) Stroma. (EN) Endothelium. (DM) Descemet membrane. (PC) Pigment cells. TB stain.

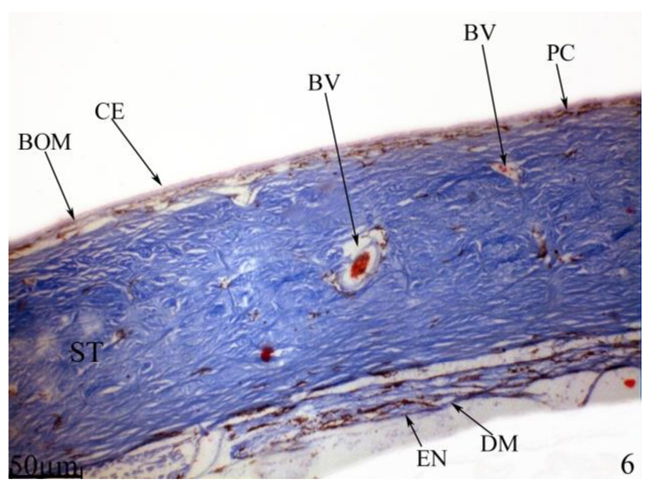

Figure 6: Cross-section in the Root of cornea. (CE) Anterior Corneal epithelium. (BOM) Bowman's membrane. (ST) Stroma. (EN) Endothelium. (DM) Descemet membrane. (PC) Pigment cells. (BV) Blood vessels. TS stain.

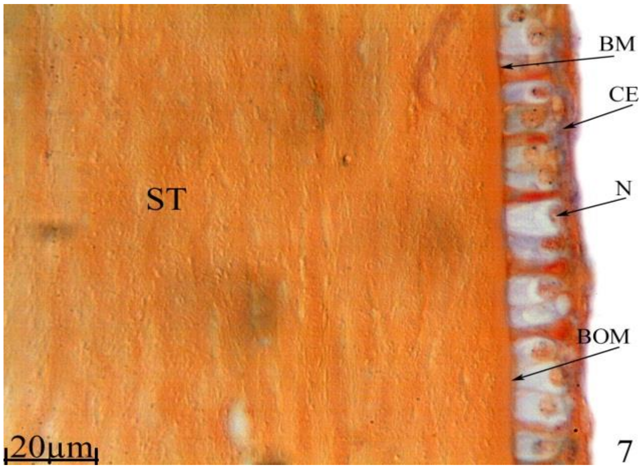

Figure 7: Cross-section in the dorsal cornea. (CE) Anterior Corneal epithelium. (BOM) Bowman's membrane. (ST) Stroma. (N) Nucleus. (BM) Basement membrane. TS stain.

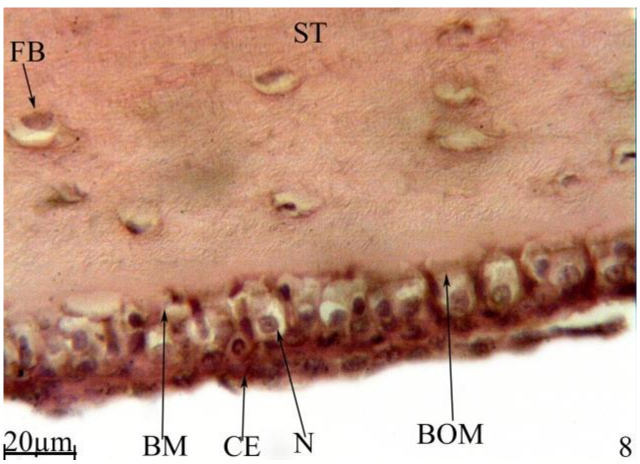

Figure 8: Cross-section in the ventral cornea. (CE) Anterior Corneal epithelium. (BOM) Bowman's membrane. (ST) Stroma. (N) Nucleus. (BM) Basement membrane. (FB) Fibroblasts. PAS stain.

Bowman's membrane has appeared thicker on the dorsal side than on the ventral side, and its average thickness on the dorsal side was 3.200± 0.024 µm, while its width on the ventral side was 2.958 ± 0.034 µm (Figure 8). The stroma of the cornea appeared consisting of three regions, the first region composed of bundles of somewhat loosened collagen fibers that contain voids that resemble circles. The second region represents the central or central region that consists of several bundles of collagen fibers that are densely compact and free from any disintegration or spaces and fibers spread among the bundles of many fibroblasts. These fibroblasts are randomly distributed among collagen fibers, while the last region consists of bundles of somewhat loosened collagen fibers, fibroblasts not observed in this region (Figure 9). Descemet membrane appeared clearly on both the ventral and dorsal sides, and its average thickness was 2.440 ± 0.015 µm. It lined with endothelium, which is composed of simple squamous epithelial cells (Figure 10).

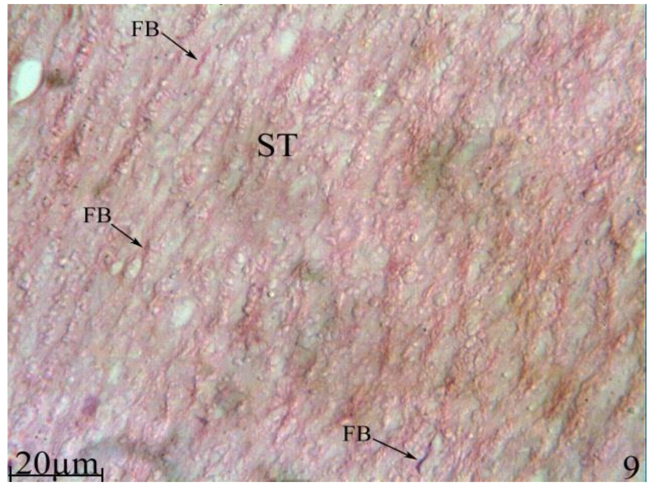

Figure 9: Cross-section in the central cornea. (ST) Stroma. (FB) Fibroblasts. H&E stain.

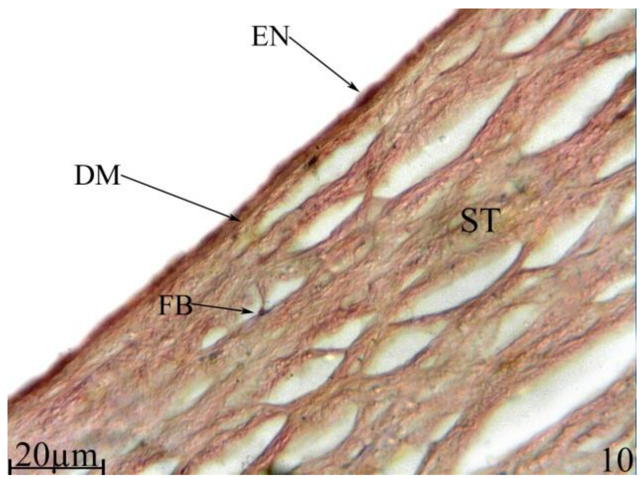

Figure 10: Cross-section in the central cornea. (ST) Stroma. (EN) Endothelium. (DM) Descemet membrane. (FB) Fibroblasts. H&E stain.

Discussion

The cornea appeared convex and made up of two anatomical regions: the peripheral area (or the corneal root), and the central part (the central cornea). The results showed that the two regions differ in thickness, as the central cornea is thicker than the peripheral cornea (the cornea root). It is a common condition in amphibians, and most birds include Corn Crake (17), *Falco tinnunculus* and *Streptopelia decaocto*, guinea fowl and Fischer's lovebird. Contrary to what appeared in the Pin-tailed Sandgrouse (18) and falcon (19) in which the peripheral cornea is thicker than the central one. The central cornea also seemed thicker on the dorsal side more than the ventral side, as it appeared on guinea fowl and Fischer's lovebird, as well as in the *Anableps anableps* and the Zebrafish.

The results showed that the cornea root contains separate clusters of pigment cells. The pigment cells are protective cells from bright light, as they reduce scattered light and prevent the colour from fading, and this leads to enhanced image contrast and visual acuity. These pigment cells also absorb short wavelengths that damage the tissue structure (20). The presence of these cells mentioned in many birds, including Turkey, tailed Sandgrouse (18) and Corn Crake (17).

The results showed that the cornea with its central and peripheral parts consists of five layers and it is a common condition in fish, amphibians, reptiles and the rest of the vertebrates. The first layer consisted of stratified squamous epithelial tissue composed of four rows of cells. The increase in the number of rows of epithelial cells has been explained to help absorb ultraviolet rays, or that the increase has significance in vision in righteousness. It is also essential to regulate the osmotic pressure because the more the layer of cells is able to reduce this pressure (21).

The results showed a difference in the thickness of the Bowman membrane between the dorsal and the ventral parts of the cornea. The presence of Bowman's membrane gives additional strength to the cornea and may act as a regulator of the movement of salts and water across the cornea (22). Also, the development of this membrane during fetal growth and development at the level of the chordates division has not been known yet clearly.

The results showed that the stroma consists of three layers of collagen fibers, is distinguished by their presence of fibroblasts. The presence of the fibroblasts indicates the animal's ability to repair the cornea when damage occurs, as well as increasing transparency in the optical path. The Descemet membrane was clear. It functions in the wild vertebrates to distribute tension and prevent general deformation of the cornea (20). It lines the inside of a layer of endothelial tissue, and this layer has an essential role in preserving the cornea from bloating as a detestable barrier to water and other pumping operations.

Among the important results that appeared in the root of the cornea of this bird, is the presence of blood vessels distributed geometrically, and this case was not previously recorded in birds except in one case and is at the root of the cornea of the seagull. The presence of these vessels may be due to the need of the cornea in this bird for high blood supply because of the dependence of this bird on the eyes to a great extent in the detection of prey. The presence of these blood vessels than increases the efficiency of the cornea in the performance of its visual functions. On the other hand, the presence of these vessels near corneal root is necessary in order not to affect the transparency of the central cornea of this bird.

Conclusions

This study concludes that the cornea in this bird is similar to that of other birds. However, it was distinguished by many specialties to meet the visual needs of this bird. It relies heavily on vision for food through its dependence on hunting as well as its adaptation to the environment.

Acknowledgments

The author is very grateful to the University of Mosul, represented by Professor Dr. Qusay Kamal Al-Din Al-Ahmadi. The Deanship of the Education college for Pure Sciences for their provided facilitates helped improve the quality of this work.

Conflict of interest

The authors declare that there are no conflicts of interest regarding the publication of this manuscript.

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التركيب التشريحي والنسجي للقرنية في باشق العصافير *Accipiter nisus*

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الخلاصة

أجريت الدراسة الحالية للتحري عن التركيب التشريحي والنسجي لقرنية باشق العصافير *Accipiter nisus* باستخدام المجهر الضوئي وعدد من الصبغات النسجية. بينت النتائج أن القرنية تتألف تشريحياً من جزأين هما القرنية المحيطية والقرنية المركزية اللذين يكونان متباينين في السمك بين مناطق القرنية المختلفة. تتألف القرنية بجزئيتها من الناحية النسجية من خمسة طبقات. تتألف الطبقة الأولى في القرنية المركزية من نسيج ظهاري حرشفي مطبق غير متقرن مكون من أربعة صفوف من الخلايا. في حين ظهر غشاء بومان واضحا وأكثر سمكاً في الجهة الظهرية منه في الجهة البطنية. أما سداة القرنية فتتكون من الياف غراوية تترتب في ثلاثة طبقات ثانوية داخل السداة ويختلف تراصها بين هذه الطبقات. كما امتازت سداة القرنية باحتوائها على خلايا مولدة للألياف تنتشر بصورة عشوائية بين الألياف الغراوية. أما بالنسبة لغشاء دسمت فقد ظهر واضحاً ومتساوي في السمك بين الجهتين الظهرية والبطنية. النسيج الظهاري المبطن ظهر بشكل نسيج حرشفي بسيط تتراص خلاياه بشكل غشاء سميك. أما بالنسبة للتركيب النسجي لجذر القرنية فظهر مشابه لتركيبها في القرنية المركزية إلا أنها امتازت بوجود خلايا صباغيه منتشرة في سداة جذر القرنية والتي تختفي في القرنية المركزية. ومن النتائج المميزة التي ظهرت في هذه الدراسة وجود أوعية دموية منتشرة في جذر القرنية. استنتجت هذه الدراسة أن التركيب التشريحي والنسجي للقرنية امتاز ببعض الصفات التي تسهم في زيادة الفعالية الوظيفية للقرنية لهذا الطائر.