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Histological Structure of the Sclera in Three Species of Birds which Differ in Nutrition Nature

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***Abstract*—** The present study dealt with the histological structure of the sclera in three species of birds that differ in their feeding, environmental, and classification. The birds were the sparrowhawk *Accipiter nisus* (carnivorous), the starling *Sturnus vulgaris* (Omnivorous), and zebra finch *Taeniopygia guttata* (granivorous). By using light microscopes, as well as the histological sections, had been stained with five different stains. The results of the sclera showed that it composed of collagenous fibers and hyaline cartilage with internal and external membranes the inner membrane appeared that they lined with pigment cells that differ in their density between the three species. The sclera appeared developed in the three species, but it more developed in the sparrowhawk. The most important results that appeared in this tunica, that the cartilage in both sparrowhawk and starling were separated in some parts of the eyeball by collagenous fibers and appeared the cartilage in sparrowhawk separated in some parts of the eyeball by blood vessels, as well as the cartilage contained pores which interspersed by blood vessels. While in the zebra finch, the cartilage was appeared in some regions separated by the blood canal. The cartilage ossifies in its connection area with the cornea in the sparrowhawk to form sclera bones, which not observed in the other two species. The cartilage also ossifies near the optic nerve to form optic nerve bone in both sparrowhawk and starling these not found in the zebra finch.

Keywords— *Sclera, Sparrowhawk, Starling, Zebra finch.*

I. INTRODUCTION

Birds have eyes that can distinguish between distances well so as not to collide with trees and other objects, and this is evident in the center of sight that forms half of the brain of birds. Also, each bird has its fingerprint, which is its eye, and when you look at it, it can never be the same in another bird because it is like a fingerprint that can never be the same [1].

The eye in the vertebrates consists of three main components: the optic components, which include the cornea, lens, and iris, the photosensitive layer, the retina, which consists of a high-capacity neural network that processes nerve signals electronically, and these signals originate mainly from the photoreceptors in the retina. This Visual pigment absorbs the light as well as the Protective casing consisting of the Pigmented choroid and the Retinal pigment epithelium. Functionally, these three parts are similar to the modern photographic camera [2].

The sclera represents the outer layer of the eye. It serves as a robust and rigid structure that preserves the shape of the eyeball and protects it from bruises. Also, it keeps the shape of the eyeball from the impact of internal and external pressure [3], and at the same time helps the sclera to make eye movement under control The brain, as it is the center of bonding of the eye muscles [4].

The sclera made up of different types of vertebrates from two parts: the outer part is the Fibrous layer, and the inner part is the hyaline cartilage [4]. Numerous studies and researches that deal with sclera in birds indicate that they are similar in structure to other vertebrates' sclera.

The sclera of birds in their contact area with the cornea may contain bone structures known as scleral bones. These bones differ in their number according to different types of birds, as the number in most birds' ranges between 10-18 bones, and the presence of these bones has an essential role in maintaining the spherical shape of the eye [5].

II. METHODOLOGY

The birds (five birds of each type) were anesthetized with Chloroform, after which the head decapitated. Feathers, lower jaw, and pilgrims' bone cut and the eyeball was removed from the Orbit quarry using fine tweezers. The histological sections prepared according to the method of [6]. Several histological stains were used in this study to learn more about the structural details of the sclera. These stains were hematoxylin-eosin (HE), Periodic acid–Schiff (PAS), Triple Stain (TS), Toluidine blue (TB), and Alcian blue (AB).

III. RESULTS AND DISCUSSION

The results showed that the sclera layer in sparrowhawk is well-grown and is composed of several rows of collagen

fibers. They differ in thickness from one area to another in the eyeball as they appeared in the form of a thick layer of collagen fibers in the middle of the eye. Their thickness ranged between (140.752-312.022 mm), and they are of somewhat disjointed rows and separated from each other (fig 1A). At the same time, it appeared in a relatively thin layer on the dorsal side near the optic nerve, the average thickness of which was approximately (139.752mm), and these fibers are highly compact (fig. 1B). These fibers interspersed with pigment cells distributed unevenly in the different areas of the sclera. They may found sparsely, and this is what appears along the sclera. Moreover, they appeared in continuous rows surrounding the collagen fibers from the outside, especially in the ventral region of the eye. In contrast, they appeared in other areas, and intermittent rows are surrounding the collagen fibers (fig 1A, C). Inside to the collagen fibers, there is hyaline cartilage surrounding the eye from the optic nerve at the back of the eye to the frontal area at the beginning of the corneal contact with the sclera in the frontal area of the eye (fig 1A, B, C).

However, this cartilage does not continue in some areas of the eye, as it found that there is a network of blood vessels belonging to the uvea layer that separates this cartilage into two parts on the dorsal temporal side. It is one of the essential and distinctive results that appeared in this study (fig1D). It also found that this cartilage is separated from one another in the ventral region close to the optic nerve, as the two parts of the cartilage are separated by a layer of collagen fibers affiliated with the sclera (fig 1E). The thickness of the cartilage varies from one region to another, as it appeared relatively thick on the front side, and its thickness approximately (109.186mm). At the same time, it was relatively thin in the middle region of the eye, as its average thickness was around (84.925mm) (fig 1A).

Among the essential results that appeared in this study is the presence of pores in some areas of cartilage through which the blood vessels enter from the uvea layer to the sclera and line a number of these pores with pigment cells (fig 1E, F). Also, among the important and distinctive results of the sclera layer of this bird is that the collagen fibers that separate the cartilage in the area near the optic nerve extend to the inner side of the cartilage so that the cartilage surrounded by the collagen fibers from the inner and outer sides, and the cartilage in this region also contains a series of pores that penetrate it. This condition not previously observed in studies dealing with the sclera (fig 1E, F). Also, from the important results that appeared are the presence of areas in the cartilage where there is a decrease in the fossil-like area in which the choroid expands, and the blood vessels enter only in this decrease while the retina does not enter in it, and this condition is recorded for the first time as far as we know. The inner and outer perichondrium appeared obviously in most regions and not clear in some areas (fig 1A, F). Hyaline cartilage is also lined with a layer of melanocytes that appear continuous row or as a membrane in some scleral areas (fig 1A). Also, from the important results that also appeared in the sclera of this bird is the ossification of a portion of the cartilage on the dorsal and ventral side near the optic nerve, which is called the optic nerve bone (fig 1E), other bone areas called sclera bones appeared in the front area of the eye (fig 1G).

The sclera in the starling appeared similar to the sclera in sparrowhawk from the basic structure, as it consists of a layer of collagen fibers that are more compact than it looked in sparrowhawk in most areas of the eye. In contrast, it appeared in other areas less dense (fig 2A). Asymmetric thickness seemed thicker on the side of the middle of the eye, and its average thickness (94.144mm), while it appeared less thick on the back, as its thickness (37.686mm)(fig 2A). Among the critical results that appeared in the sclera of this bird is the presence of muscle fibers permeating the collagen fibers in some regions (fig 2A, B). A thick muscle layer was observed surrounding the collagen fibres from the outside and attached directly to them from the sides of the middle of the eye (fig 2C).

The collagen fibers interspersed with melanocytes, but they are much less prevalent than in sparrowhawk, as these cells appeared very little within the collagen fibers, and these cells may disappear from the fibers permanently in some of the areas of the sclera. Still, in the surroundings of collagen fibers, these cells appeared sporadically. The hyaline cartilage seemed less thick than it is in the sparrowhawk, but the chondrocytes and their lacunae are more numerous and more compact than their counterparts in the sparrowhawk (fig 2A). The thickness of this cartilage varies in the posterior regions of the sclera, as it was thick in the area near the optic nerve, its average diameter was approximately (154.185mm)(fig. 2D). In contrast, the lowest thickness appeared in the ventral temporal region, and its average depth(32.070mm) (fig 2C).

The sclera of this bird contains an area similar to what appeared on it in the sclera sparrowhawk, as cartilage seems to be separated by collagen fibers, and did not appear continuously in the dorsal-ventral region (fig 2E). This separation area appears more extensive than it looked in sparrowhawk. The perichondrium of the hyaline cartilage did not appear visible as it appeared in the sparrowhawk. Whereas the inner perichondrium appeared clearly and thicker than it appeared in the sparrowhawk. Its average thickness (3.048mm) (fig 2A). It lined toward the uvea with a continuous layer of melanocytes that appear in a thicker than appeared in the sparrowhawk (fig 2A).

On the other hand, there are pores in the sclera of this bird that appeared in the cartilage as in the sparrowhawk and lined with blood vessels. Still, these pores appear wider than in the sparrowhawk and are of an elongated oval shape, and the blood vessels appear wider than in the sparrowhawk (fig 2F). Also, the optic nerve bone appeared in this bird as well, and it is more visible and broader than in the sparrowhawk, and this bone appeared in the cartilage

on both sides of the optic nerve, while the scleral bones not observed in this bird (fig 2G).

The sclera in the zebra finch was less developed, and growth than in the other two birds, and collagen fibers appeared highly compact. They did not show any disintegration, in contrast to what mentioned in the previous two types, as it seemed about one thickness along with the eyeball, as the average width reached (16.218mm) (fig 3A, B, C). Pigment cells are also considerably less numerous than in the other two birds and appear sparsely and rarely distributed between the collagen fibers. These cells did not seem connected as they appeared in the other two types (fig 3D).

The hyaline cartilage appeared less thick than it was in the previous two types, as it seemed about one thickness along the circumference of the eye, and its average thickness (27.438mm) (fig 3A, B). Still, it is in the area near the root of the cornea in the dorsal temporal region is very thick, and the chondrocytes are smaller than the rest of the areas. The average thickness of this cartilage reached (127.690mm) (fig 3C, D). The hyaline cartilage appeared continuously from the beginning of the optic nerve to the frontal part of the eye, in contrast to what appeared in the previous two types (fig 3A, B). The chondrocytes appeared in this bird in a large and compact more than what appeared in the other two birds. The lacunae appeared to be large and occupy most of the interstitial material of the cartilage (fig 3B, C), but, in the dorsal temporal region. It found that the cartilage separated from on the other side, a blood canal connected to the blood vessel outside the sclera on the one hand, and the choroid vessels of the choroid on the other side inserted in the separation area. This case differs from what it appeared in the sparrowhawk sclera in that it contains a blood canal here while in the sparrowhawk, it includes a blood vessel, and this case recorded for the first time in studies that dealt with birds as far as we know (fig 3E).

On the other hand, the perichondrium of the cartilage appeared similar to its counterpart in the starling, as it did not appear, while the inner perichondrium did not appear in contrast to the previous two types (fig 3A), but, in some areas of the sclera, the perichondrium is visible. The contents are combined hyaline cartilage with the collagen fibers of the sclera strikingly (fig 3C). The cartilage of sclera in this bird was distinguished by lining from the inside with a thick and continuous membrane of the melanocytes thicker than it was in the previous two types. Its average thickness was (4.268mm) (fig 3B, C, D). Whereas there is no optic nerve bone and the scleral bones notice in this bird. However, one of the critical and striking results of the sclera in this bird's is the presence of a bone region on the side of the cartilage in the dorsal temporal region in which the cartilage is thick near the root of the cornea, and this bone is the spongy type (fig 3D, E, F).

Discussion

Numerous studies that have indicated that the sclera of the eye in birds is composed of two layers, the outer layer represented by the collagen fiber layer, while the interior layer represented by hyaline cartilage [7], and this corresponds to the current study.

The sclera appeared different in its thickness between the three types of birds and is thicker in the sparrowhawk than the other two types, and this is consistent with the findings of some previous studies [8],[9]. The results showed that the layer of collagen fibers varies in thickness between the three types, it appeared thicker in the sparrow then the starlings, but in the zebra finch, it appeared with about one width. This layer includes pigment cells that differ in their distribution and intensity between the three types, as they are denser and organized in sparrowhawk than the other two types, and this is consistent with some previous studies [8],[10]. Al-Robaae (2010) mentioned that increasing the thickness of the collagen fibers gives the eyeball added protection.

Whereas, the cartilage appeared thicker in the sparrowhawk than in the other two types. However, it appeared in the zebra finch with almost one thickness, while it appeared in the starling in the posterior region near the optic nerve, very thick. The cartilage is surrounded on both sides by a cover known as the perichondrium. The outer perichondrium lies towards the layer of collagen fibers, which appeared clearly in the sparrowhawk, while it did not appear clearly in both the starling and the zebra finch. The inner perichondrium appeared clearly in both sparrowhawk and starling. In contrast, it did not appear in the zebra finch, the presence of cartilage in the sclera of the eye, and its thickness variation is common in birds (Abd and Abd al-Majid, 2010). We believe that the arrangement of the compositions above helps the sclera in performing its functions more efficiently in a way that suits the bird's life and environment.

The results of the current study also showed that the cartilage in the three types lined from the inside with a thick layer of pigment cells, which appeared thicker in the zebra finch of the other two types. This is similar to what appeared in the cartilage in other birds [10],[12].

The results of the study showed that the sparrowhawk sclera contains bone structures in the area of its connection to the cornea, and these bones are called sclera bones that have not observed in the other two types. These bones are essential in adapting the cornea, maintaining the shape of the eye, and protecting it from pressure changes [13].

The results of the study also showed that the cartilage in both sparrowhawk and starling ossified near the optic nerve, creating an optic nerve bone that not observed in the zebra finch, and this is consistent with what was stated [14]. It believed that the function of the optic nerve bone is to protect the optic nerve from pressure [9].

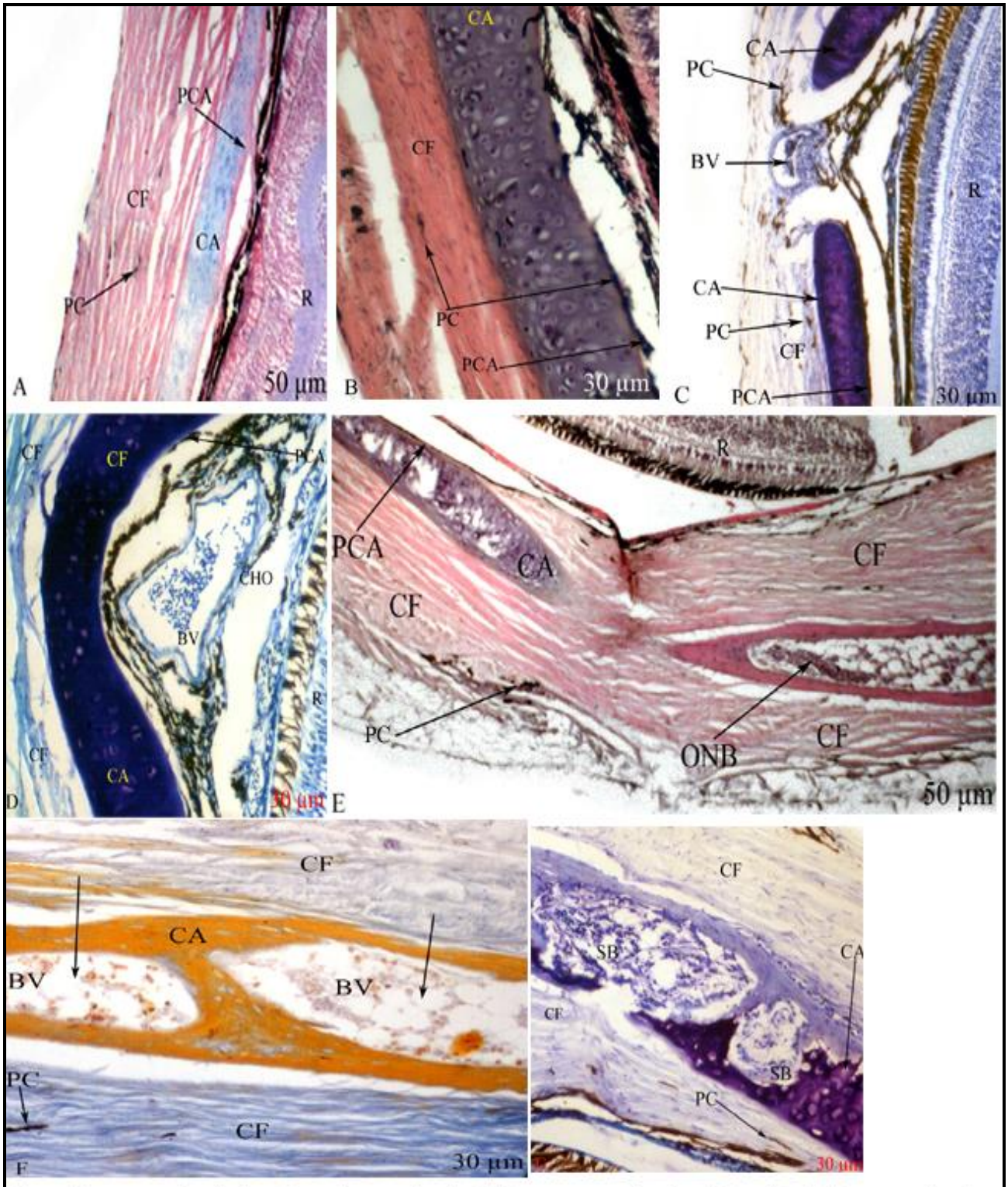

Figure (1) cross-section in the sclera of sparrowhawk; (A) Dorsotemporal region (AB stain); (B) Dorsonasal region (PAS stain); (C) Dorsotemporal region (TB stain); (D) Dorsotemporal region (TB stain); (E) Ventrotremporal region (H&E stain); (F) Dorsotemporal region, the arrows refer to the pores in cartlage (TS stain); (G) Dorsonasal region (TB stain). PC pigment cells ;PCA perichondrium ; CF Collagen fibers ; CA cartlage ; R retina ;BV blood vessels; CHO Choroid ; ONB Optic nerve bone; SB Scleral bones .

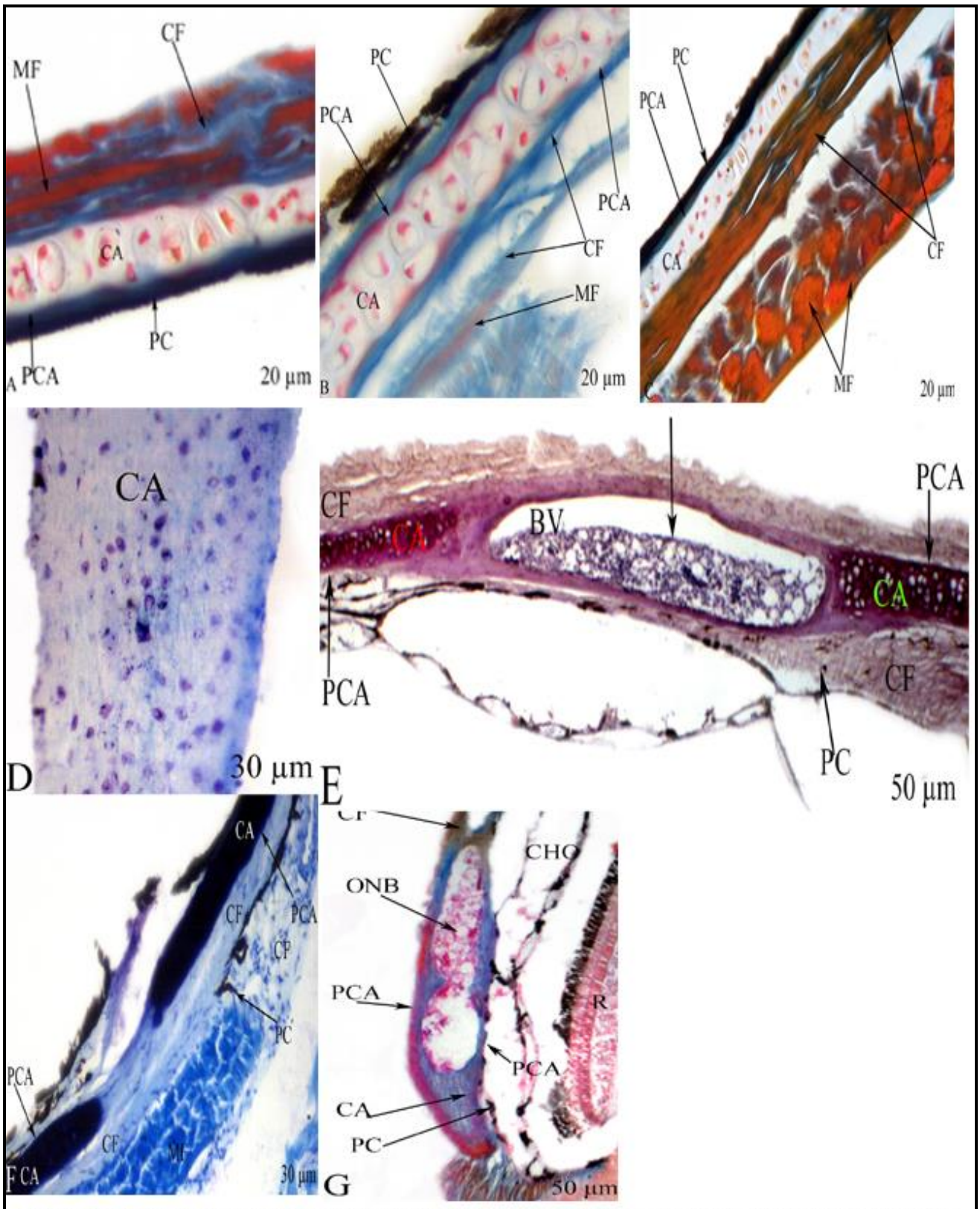

Figure (2) cross-section in the sclera of Starling; (A) Ventronasal region (TS stain); (B) Dorsotemporal region (TS stain); (C) Dorsonasal region (TS stain); (D) Ventrotremporal region (TB stain); (E) Ventrotremporal region (H&E stain); (F) Ventronasal region, the arrows refer to the pores in cartlage (TB stain); (G) Dorsotemporal region (TS stain). PC pigment cells ;PCA perichondrium ; CF Collagen fibers ; CA cartlage ; R retina ;BV blood vessels; CHO Choroid ; ONB Optic nerve bone; MF Muscle fibers .

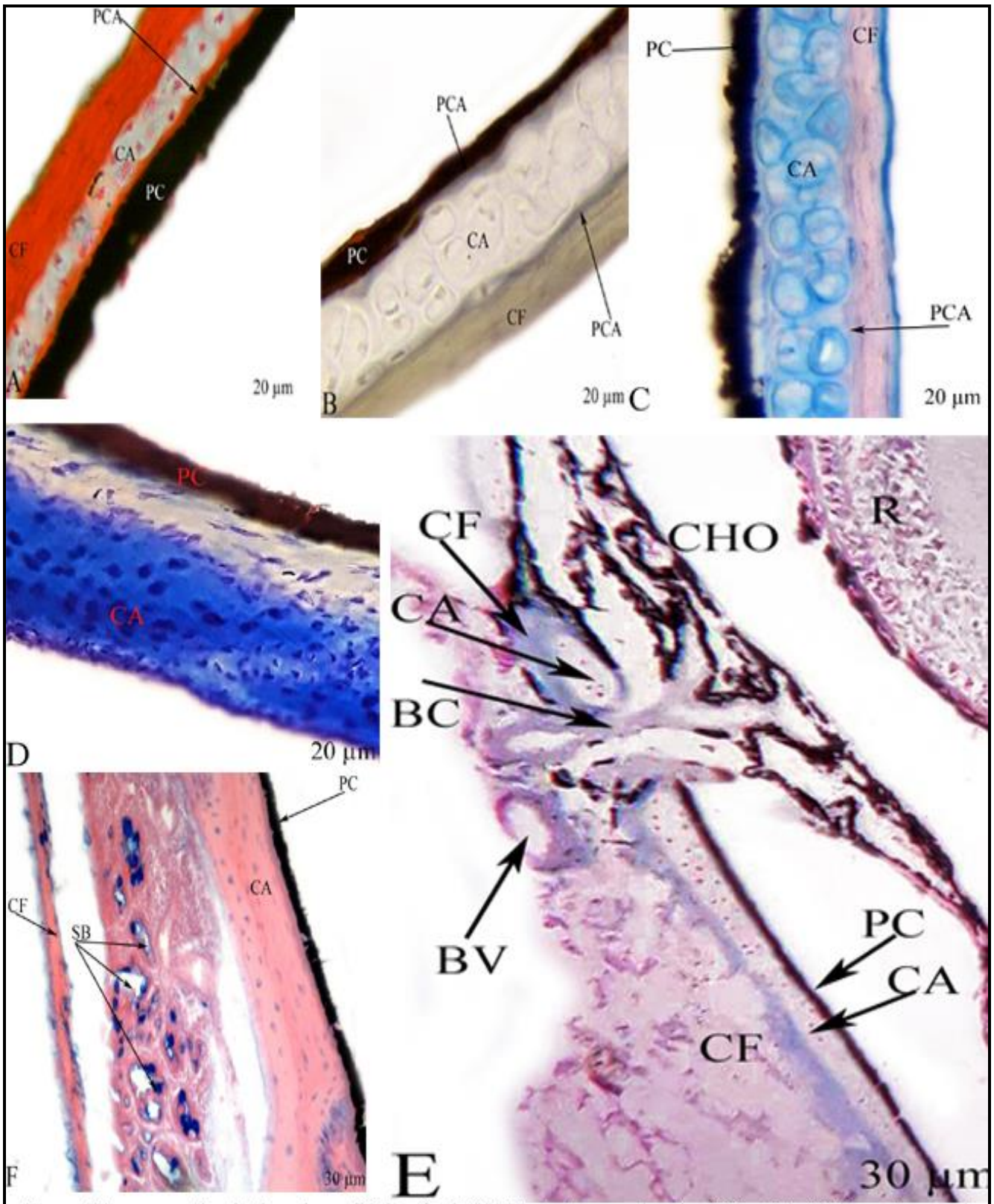

Figure (3) cross-section in the sclera of Zebra finch; (A) Ventrotremporal region (TS stain); (B) Dorsotemporal region (TS stain); (C) Ventrotremporal region (AB stain); (D) Dorsonasal region (TB stain); (E) Dorsotemporal region (TS stain); (F) Ventrotremporal region (AB stain). PC pigment cells ;PCA perichondrium ; CF Collagen fibers ; CA cartlage ; R retina ;BV blood vessels; CHO Choroid ; SB Scleral bones; BC blood canal.

Among the critical results that appeared in the sclera of sparrowhawk and starlings are the presence of pores in some areas of the cartilage through which the blood vessels enter from the uvea layer to the sclera and these pores lined with pigment cells. It believed that they have a role in increasing the scleral blood flow, and this leads to improved nutrition. Also, from the distinctive results that appeared in the sclera of sparrowhawk are that the cartilage occurs in a fossil-like drop. This decrease occupies a network of blood vessels belonging to the choroid. We believe that this result is recorded for the first time as far as we know, as this result not observed in previous studies, and this region may work with the blood vessels that it contains increases eye and retinal nutrition by increasing the surface area in this region.

Scleral cartilage has also appeared in sparrowhawk discontinuous, as it is separated by a network of blood vessels belonging to the uvea layer and in other areas, it is separated by collagen fibers that extend to the inner side of the cartilage, and thus the cartilage becomes surrounded by collagen fibers from the inner and outer sides. The separation of cartilage by the collagen fibers also appeared in the starling sclera; this result not observed in previous studies. Therefore, this region may work to increase the strength and durability of the scleral cartilage by increasing the linkage between its parts. It also gives flexibility to the sclera in the face of the pressures it exposed to during the flight.

Also, the cartilage appeared in the sclera of the zebra finch also separated by a blood canal connected to a blood vessel located outside the sclera on the one hand and the vessels of the choroid, on the other hand, this condition is recorded for the first time as far as we know. It believed that these blood vessels contribute to increasing the blood nutrition of this region of the eye.

IV. CONCLUSION AND FUTURE SCOPE

We can conclude from the results of the current study that the composition of the sclera is essential in preserving the eye in birds, especially during flight. The more the bird relies on feeding it on a moving food, the composition of the sclera becomes more complicated to suit all conditions when obtaining food. It pronounced in the sclera of the sparrowhawk.

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