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Morphological and Histological Study of the Tongue in Slender-Billed Gull (*Larus genei*)

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Abstract

All birds are adapted to their different environments with respect to food sources, reflecting their different life styles, therefore the birds have different feeding habits, prominent differences in the structures of their tongues. The study aimed to identify the morphological characteristics and histological structure of the tongue in Larus genei and its relationship to the feeding style. The study used five birds, which were purchased from Al-Ghazel local market in Baghdad prominence. The animals were anesthetized, and the tongue was removed and studied morphologically. The tongue was fixed using Formalin (10%) for 48 hours, and the histological sections were prepared and stained using Hematoxyline-Eosin for the histological study .The results showed that the tongue in *Larus genei* characterized by the presence of a median sulcus, and the conical papillae are spread on the superior surface of the tongue. The superior and inferior surfaces of the tongue are covered with stratified squamous epithelium. The epithelial lining the lamina propria, which composed from loose connective tissue; in addition to, the tongue is supported by a structure made up of hyaline cartilage. For the longitudinal muscularis layer, it appears thin and occupies a small part compared to the rest of the layers. Conclusion: The study revealed that *L. genei* tongue possesses morphological characteristics such as conical papillae and a median groove; it was found the presence of hyaline cartilage.

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1. INTRODUCTION

Many researchers have studied the digestive system carefully, and the tongue is one of this system parts, which plays an important role in all vertebrates [1-3], Thus, the recent study was intended to study the tongue of *Larus genei*. The bird is a seasonal wild bird that inhabits the north of the Old Polar Region and the west of the New Polar Region. In addition to, it is a winter visitor bird and is common in Egypt, the Arabian Gulf, and Kuwait. It belongs to the Laridae family, of the Charadriiformes order, and appears to be medium in size, white in color, and has a long, thin and red beak [4]. Several studies have examined the structure of the tongue in many birds including *Gallus domesticus* [5], *Columba livia gaddi* Gemlin, 1789 [6] and Peregrine falcon (*Falco peregrinus*) [7]. The birds have adapted to live in different environments, which have led to the formation of their tongues in different shapes to suit their lifestyles and the nature of nutrition. The tongue is generally divided in most animals into three parts: apex (A), body (B), and root (R) [1], [8-10]. The tongue in all birds contains lingual papillae that function like teeth, for crushing the solid food (grains) [11]. Interestingly, the tongue of birds lacks internal muscles and taste buds, and it appears covered with a keratinized substance and contains pointed protrusions at its root. The tongue of some birds is split at its free end, whereas in other species it looks more like a brush. While in the parrot, it appears thick due to the presence of fats and blood vessels, whereas the woodpecker can extend its tongue a long distance to catch insect larvae from tree trunks [12].

The tongue is covered with a squamous epithelium, and the tongue of most birds contains lingual glands, while the glands are absent in others as in pelicans [13]. The aims of study were identified the morphological characteristics and histological structure of the tongue in Larus genei and its relationship to the feeding style.

2. MATERIAL AND METHODS

The study conducted according to instructions and regulations of animal ethics of University of Baghdad College of Education for Pure Science (Ibn Al-Haitham). The study included the collection of five males' samples of *L.genei* birds at age 4 years, which purchased from Al-Ghazel market in Baghdad provenance and classified in the Iraqi Natural History Museum, Department of Vertebrates. The birds were transported to the comparative anatomy laboratory at the College of Education for Pure Sciences (Ibn Al-Haitham), and then the morphological description of the tongue was studied. The birds were anesthetized by using chloroform in a killing jar containing cotton soaked with chloroform and left for (5-7) minutes until the bird stopped moving. The tongue was removed by making an incision on both sides of the beak and removing the tongue by cutting the frenulum from the inferior surface of the tongue and continuing backwards until it reached the end of the tongue. The tongue placed in a normal solution for the morphologically study using a dissecting microscope and images were collected using a Xiaomi 11 mobile camera with a resolution of 108 megapixels. The other samples were fixed directly using formalin (10%) for 48 hours, and the histological sections were prepared according to the method of [14, 15] which included dehydration, clearing and embedding processes. The cross sections of 5µ thickness were obtained using a rotating microtome. The sections were stained using Hematoxylin and Eosin stain to clarify the histological structure of the organ; the prepared slides were examined using an optical microscope (MEIJI TECHNO) equipped with an Omax camera.

3. RESULTS

a. The morphological description

The results of the study revealed that the tongue of a slender-billed gull bird appears needle-shaped and thin in the front and gradually expands towards the back with a sharp and pointed end, and reaches a length of 3 cm (Figure 1). Furthermore, the tongue is connected from its lower part to a membranous fold called the lingual frenulum (Figure 2). The tongue consists of a superior and inferior surface; the results also showed that the superior surface of the tongue contains a deep and clear groove that divides it into two halves called the median sulcus which extends along the tongue, making the tongue take a concave shape (Figure 1).

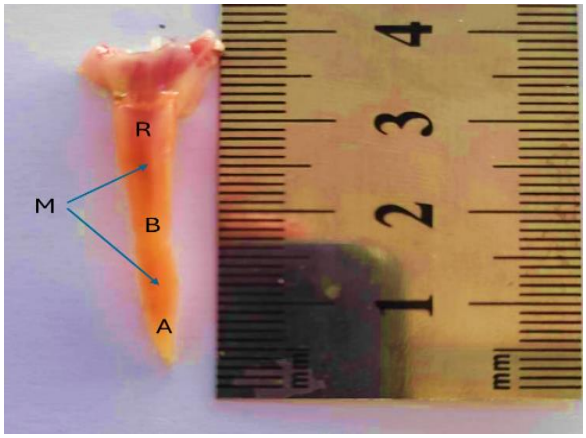

Figure 1: The Morphological feature of the tongue showed: three parts of the superior surface: Apex (A), Body (B), Root(R) Median sulcus (M).

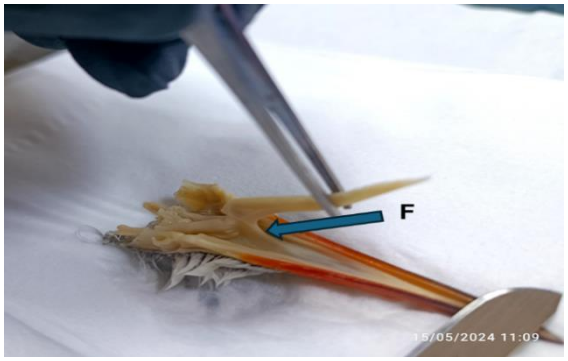

Figure 2: The Frenulum (F) in *L.genei*.

The tongue in slender-billed Gull bird is divided into three parts (apex, body, and root). The apex appears to be very thin and pointed, while the body is the longest part of the tongue and appears wider than the apex, whereas the root is the third and last part of it (Figure 1). The study also observed that the superior surface of the tongue contains rough mechanical lingual papillae represented by conical papillae, whereas the inferior surface is devoid of papillae. The conical papillae are spread along the tongue, as appear few in number, and their distribution varies between the parts of the tongue. At the apex, the conical papillae distribution is very few and small in size, while they appear larger in size and more widespread in the area of the lingual body. As for the root, it has been shown that there are large-sized conical papillae facing backwards and lined up in a V-shape (Figure 3).

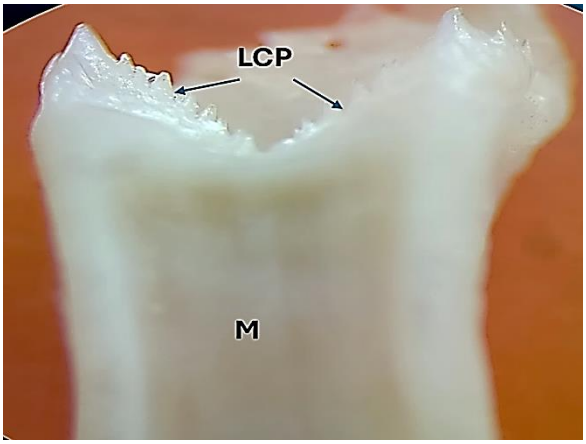

Figure 3: The Root region of *L. genei* tongue showing: Large conical papillae (Lcp), Median sulcus (M).

b. The results of the histological study

The microscopic examination found that the tongue of the *L.genei* bird cover of an epithelial layer composed of slight-keratinized stratified squamous epithelial tissue at the superior surface, while it is keratinized on the inferior and lateral surfaces of the tongue. The keratinization is thicker on the inferior and lateral surfaces in the apex of the tongue compared to the rest of the tongue parts, as the keratinization begins to decrease toward the root (Figure 4 and 5).

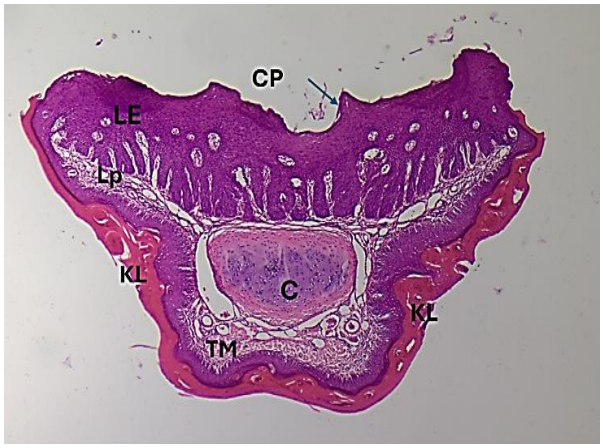

Figure 4: A Cross section in lingual Apex of *L. genei* showing: Conical papillae (CP), Lining epithelium (LE), lamina propria (Lp), tunica muscularis (TM), cartilage (C), keratinized layer (KL), (H&E) (10X).

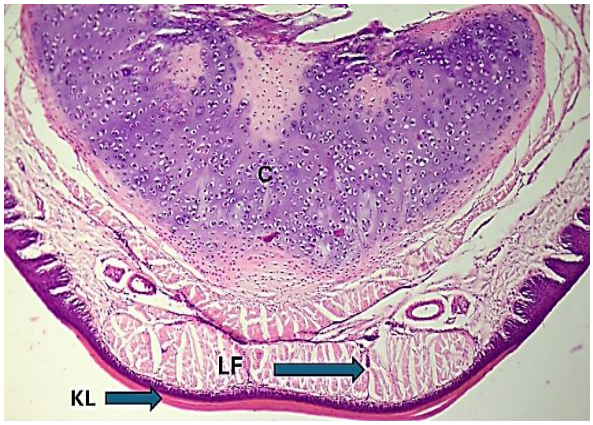

Figure 5: A cross section in lingual body of *L. genei* showing: longitudinal fibers (LF), keratinized layer (KL), Cartilage (C), (H&E) (10X).

Furthermore, the lingual papillae, represented by conical papillae, appear in this layer (Figure 6).

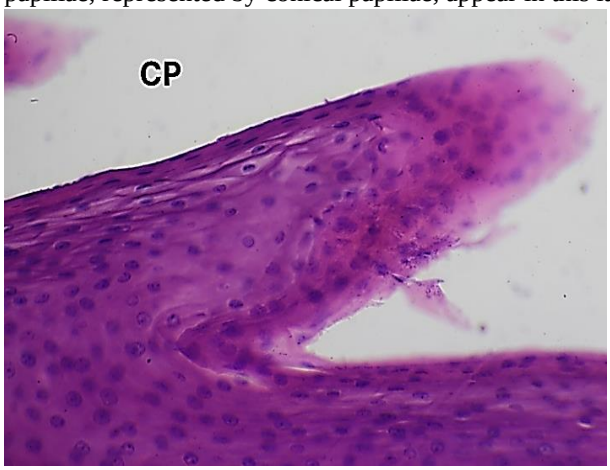

Figure 6: A longitudinal section in conical papillae (CP) of the lingual root in *L. genei*, (H&E) (40X).

It has been shown that the tongue in the sample of the study lacks the taste papillae and taste buds. Moreover, the findings of this study found that the basal layer generating the epithelial lining consists of a single row of columnar or cuboidal cells with oval-shaped nuclei arranged at different levels located on the basement membrane (Figure 7).

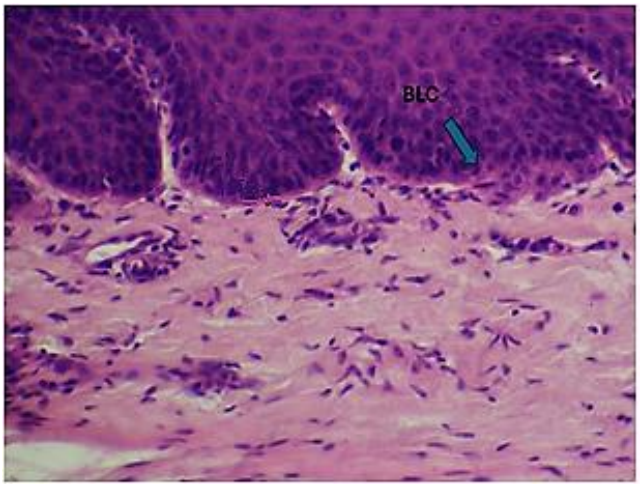

Figure 7: A longitudinal section in lingual body of *L.genei* showing: basal layer cell (BLC), (H&E) (40X).

The epithelial lining is followed by the lamina propria, which is composed of loose connective tissue. In this layer, the lingual glands are found, which are located in the lingual root. Additionally, these glands are of the mucous type, composed of low columnar cells and a relatively large and clear cavity; the nuclei are compact, basal, and dark in color (Figure 8).

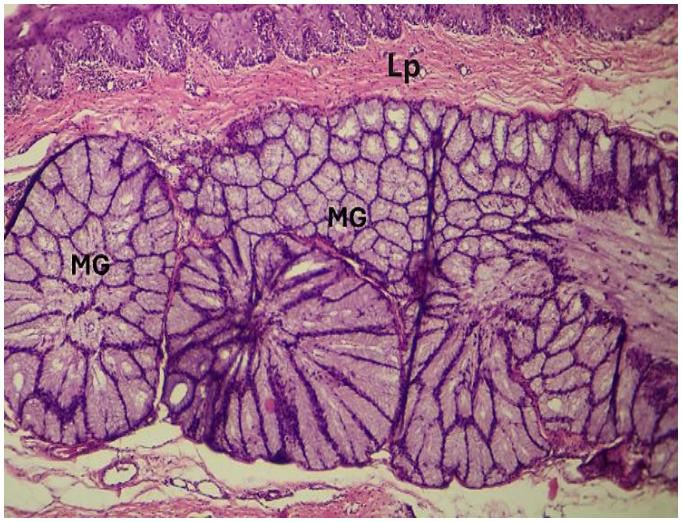

Figure 8: A longitudinal section in lingual root of *L. genei* showing: Lamina propria (Lp), mucous gland (MG), (H&E) (40X).

The microscopic examination revealed that a cartilage structure consisting of hyaline cartilage that extends along the tongue and represents the supporting structure of the tongue (Figure 4 and 5). Whereas the longitudinal muscularis layer appears as a thin layer that occupies a small part compared to others layers; it consists of longitudinal muscle fibers arranged in bundles; it was also found that there was no fatty tissue between the bundles of muscle fibers (Figure 5).

4. Discussion

a. The Morphological description

The results of the study observed that the tongue of *Larus genei* appears thin, needle-shaped, with a sharp and pointed end [4]. It is thin in the front and gradually expands towards the back, and its length reaches 3 cm. In addition, the tongue is connected from its lower part to a membranous fold called the lingual frenulum, and this result is in agreement with the findings that found in most of the previous studies on the tongue of birds [6],[16], respecting the difference in the shape of the tongue in different types of birds, which may be due to the nature of the bird's diet and lifestyle.

Interestingly, the tongue consists of a superior and inferior surface; the superior surface contains a deep and clear groove that divides the tongue into two halves, called the median sulcus which extends along the tongue, making the tongue take a concave shape. This result is consistent with previous studies that studied the common quail, domestic pigeon and chucker partridge and whose tongues have a median sulcus that divides the apex and body into two halves [17]-[20]. On the other hand, the result of this study contradicts other studies in that found the tongue of the studied birds lacks a median sulcus [16], [20]. The variation in the presence or lack of the median sulcus may be due to the type of bird's environment and diet. It is believed that the function of the median sulcus is to regulate the path of food towards the esophagus [19].

The tongue in ***L.genei*** is divided into three parts: apex, body, and root. The apex appears to be very thin and pointed, while the body is the longest part of the tongue and appears wider than the apex, whereas the root is considered the third and last part of the tongue. This result in agreement with previous researches [6], 13], [21].

The lingual papillae are spread along the tongue. It is revealed through visual examination that the present type of papillae is only with conical papillae, as they appear few in number and their spread are varies between parts of the tongue. At the apex, their spread is very few and small in size, while appear larger in size and more spread in the body of tongue, whereas in the root it was found that there are large conical papillae facing backward and lined up with a V-shape. The result is in agreement with many studies that studied the tongue of birds in terms of their possession of conical papillae, as the distribution of these papillae differs from one bird to another [6,10,22]. The function of the conical papillae is a mechanical function, as they work to hold on to food, push it back, and prevent it from sliding out of the mouth.

b. The Histological study

The results of microscopic examination showed that the tongue in *L.genei* bird consists of the mucous layer composed of non-keratinized stratified squamous epithelial tissue. In addition to, the keratinization appears at the inferior and lateral surface of the tongue, as keratinization is thicker at the inferior and lateral surfaces of the tongue apex compared to other parts of it, where it begins to decrease toward the root. This result is consistent with a previous study on *Sturnus valguris* tongue [13], while it contradicts the results of [23,24] that studied the tongue of *Acridotheres tristis*, as the bird's tongue was devoid of keratinization. The variation in degrees of keratinization may be due to the nature of the habitat as well as the type of food, whether it is dry or wet in nature.

Furthermore, in the mucous layer, the mechanical lingual papillae were detected, represented by conical papillae, whereas the tongue lacks taste papillae and taste buds. This finding is in agreement with study of [25], while it contradicts with the results of [26], as the bird's tongue (Merlin) contained the taste buds. The difference in results could be due to the nature of the nutrition.

The results of the study also showed that the basal layer generating the epithelial lining consists of a single row of columnar or cuboidal cells with oval-shaped nuclei arranged at different levels located on the basement membrane; this result is consistent with [6] study which appear that the tongue of *Columba livia gaddi* contains of a single row of columnar cells appeared polygonal with round nuclei. Moreover, it was found that the lamina propria composed of loose connective tissue; this result was in agreement with [7] in *Falco peregrinus* and [25]. in pigeon bird which they found that the lamina propria in *Falco peregrinus* and pigeon bird consisted of loose connective tissue containing collagen fibers, while not consistent with [6] study that reported that the lamina propria in *Columba livia gaddi* was composed of dense and irregular connective tissue.

The study revealed the presence of lingual glands in the lingual root only, and also clarified that these glands are of the mucous type, composed of low columnar cells and a relatively large clear cavity. While the nuclei are compact, basal, and dark in color, as there are no serous glands. The finding is in agreement with the result of *Colomda livia gaddi* tongue [6] and contradicts with the tongue of *Falco peregrinus* [7], because the location of the lingual glands in the tongue increases towards the lingual root [6]. The variation in the location and type of lingual glands could be due to their function in the moistening food.

Several previous studies, that studied the tongue of birds, have shown the presence of a structure that supports the tongue consisting of hyaline cartilage [6],[21], and these observations are consistent with the results of the study. In addition to, the study found that the muscularis layer is thin and occupies a small part of the tongue compared to the other layers; it consists of longitudinal muscle fibers arranged in bundles. Furthermore, it showed the absence of fatty tissue between the bundles of muscle fibers. These results are consistent with the results of [21] study as it was shown the lingual muscles in tongue *Gallus gallus* are limited to the body and root of the tongue, whereas the study showed that the muscles extend along the tongue, which was in agreement with [7] study on *Falco peregrinus* bird which shown the lingual muscle bundles are parallel to the long axis of the tongue.

However, the study of [13] appeared that the muscles in *Sturnus valguris* bird were located only in the lingual root, and this discrepancy maybe due to the importance of the lingual muscles in bird, which is determined by the nature and type of nutrition. Therefore, the muscles have an essential role in achieving different movements as well as it has a role in phenotyping and modeling the tongue.

5. CONCLUSION

The study concluded that *L. genei* tongue possesses morphological characteristics such as conical papillae (with mechanical function) and a median groove that regulates the movement of food towards the esophagus. Histologically, it was found with presence of cartilage, muscularis limited supports the tongue in addition to the keratinized layer located on the inferior and lateral surface of the tongue. These characteristics are related to the feeding pattern adopted by the bird.

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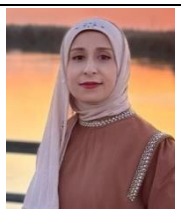
BIOGRAPHIES OF AUTHORS



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