The Egyptian Journal of Hospital Medicine (October 2017) Vol.69 (3), Page 2115-2127

Morpho-Functional Adaptations of the Lingual Epithelium of Two Bird Species Which Have Different Feeding Habits

Ali G. Gadel-Rab[1], Nahed A. Shawki[2], Samy A. Saber[3]

1- Department of Zoology, Faculty of Science, Al-Azhar University, Assuit, 2-Department of Zoology, Faculty of Science, Assuit university 3- Department of Zoology, Faculty of Science, Al-Azhar University, Cairo, Egypt

Corresponding author: Ali G. Gadel-Rab, Aligamal200992@yahoo.com

ABSTRACT

The dorsal epithelium of the free portion of tongue of laughing dove, *Streptopelia senegalensis* (granivorous) is characterized by presence of well-developed keratinized epithelium with desquamate parakeratinized one which is covering the anterior part of the free portion. Highly keratinized and stratified epithelium is covering the ventral surface of the anterior third of the free portion of the tongue and forming the lingual nail. Laryngeal area is covered dorsally by non-keratinized squamous epithelium. Frenulum is covered by a transitional-like epithelium. Both the dorsal and ventral surfaces of the free portion of the tongue of the common hoopoe, *Upupa epops* (insectivorous) are covered by thin non-keratinized squamous epithelium except that covered the areas of lingual tubercles, lingual wings and lingual papillae, which are covered by epithelium furnished dorsally by detached keratin. The laryngeal area is covered by non-keratinized squamous epithelium. The frenulum is covered by non-keratinized stratified squamous epithelium. It has been summarized that the differences in the structures of the avian tongue reflecting the differences in the feeding habits. The epithelium covering the tongue of the laughing dove is constructed for high mobility (up-down movement) in comparison with the epithelium of the common hoopoe that depends on its first action of feeding on the movement of the jaws and beak.

Keywords: laughing dove, common hoopoe, lingual epithelium, salivary glands, and adaptations.

INTRODUCTION

The feeding mechanism is an important factor that determines the success of adaptations of vertebrates to their environment and of their persistence through procreation [(1)]. During feeding process, the lingual apparatus plays a principal role, together with other organs within and near the oral cavity, in particular in tetrapods.

All birds are adapted to their different environments with respect to food resources. Reflecting their different life styles, birds have different feeding behaviors with corresponding differences in the size and structures of their lingual apparatus[(2)]. The structural adaptations of the lingual apparatus of birds vary greatly among the different species project an interesting subject for functional morphological studies and attracted the attention of many investigators[(3-7)] on the various feeding behaviors and types of food, frequent structural modifications of the lingual system (Mucosa; distribution of mechanical papillae and the area and degree of keratinization of the stratified epithelium) can be observed in birds[(8)]**.** Such modifications result in different tongue mobility and the ability to slide the tongue out, extract food and manipulate food in the beak cavity.

Columbiformes, which is avian granivorous have an elongated and narrow tongue with short and curved bill, exhibit great difference in the shape and volume of their tongues as well as the shape and volume of their bill than that of bucerotiformes which feeds on insects and possess short and broad tongues with a very elongated and straight bill. Thus, the questions are "are the lingual epithelium and its derivatives of papillae, tubercles and salivary glands in the two bird species modified to perform their action according to their feeding behavior? This question attracts the attention of the authors of the present study. The laughing dove, *Streptopelia senegalensis* (Columbifromes) and the common Hoopoe, *Upupa epops* (Bucerotiformes**)** which are sympatric species in most areas of Egypt were chosen for this investigation.

Aim of the work: the study of functional morphology of the lingual epithelium of both the laughing dove and the common hoopoe attempts to correlate the structure of the lingual epithelium of the different components with contemplate mechanical performance during the feeding process. The present study included a morphological description of the tongue of the two chosen birds as well as histological investigation and scanning

Received: 9 /8 /2017
Accepted:18 /8 /2017

DOI : 10.12816/0041069

electron microscopy (SEM) for the lingual epithelium and its derivatives of the two studied bird species.

MATERIALS and METHODS

Specimens of the common laughing dove, *Streptopelia senegalensis* and the common Hoopoe, *Upupa epops* were obtained from Abou Rawash (North of Giza, Egypt). A digital camera was used to get photos of the tongue. For light microscopic investigations, some specimens were fixed in 10% neutral formalin for two days and prepared for paraffin embedding; serial sections of 5µm were prepared and then stained with Haematoxylin and Eosin and Masson's trichrome stain[(9)]**.** For scanning electron microscopy specimens were fixed in 5% glutaraldehyde in a cacodylate buffer for 48 hr at 4ºC and washed in three changes of 0.1 % cacodylate buffer, then the specimens were post-fixed in a cacodylate buffered solution of 1% osmium tetroxide for 2 hr at 37ºC. The specimens were washed in the same buffer three times, dehydrated and then infiltrated with amyl acetate for two days. The drying of specimens was accomplished by the critical point drying using liquid Co2, mounted and sputter-coated with gold. The specimens were examined on a Jeol scanning electron microscope (J S M-5400I V), at 15 kv.
The study was approved by the Ethics Board of Al-Azhar University.

RESULTS

GENERAL MORPHOLOGY

The avian tongue is distinguished into three regions; the tongue body (Free portion, FRT), the tongue root (Laryngeal area) and the posterior paired hyoid horns (HH).

In the laughing dove, *Streptopelia senegalensis*, the tongue is elongated and narrow and drops slightly towards the bottom of the bill. The total length of laughing dove's tongue was about 3.2 to 3.5cm; it was longer than the length of the mandible which is 3cm (Figs.1a). While, the common hoopoe, *Upupa epops* possess reduced semicircular tongue. Its total length was about 2 cm shorter than the length of the mandible that is about 7.3 cm. (Fig.14).

The free portion of the tongue of the laughing dove (FRT) is conical structure with sharp-rostral tip. Dorsally, it is invaginated by a shallow longitudinal sulcus (Sulcus Lingualis,) that extends along the mid-dorsal line to its caudal margin (Figs.1a&3,8a). The caudal margin has paired lingual wing (LW), each of which ends with large postero-lateral papilla. That papilla is long, conical and pointed near its base with approximately 3cm length (Figs.1a& 9). Meanwhile, there are about 8-12 short, thick, entire, and tiny conical papillae of varying sizes (Papillae lingualis, PL) lie between the two large posterior lateral papillae and set closely to the caudal margin of the free portion of the tongue. These papillae are arranged symmetrically in single row and pointed caudally towards the laryngeal area. There are 1-2 papillae on either sides and they are bifurcated (Figs. 9& 10a, b). Ventrally, the free portion of the tongue is semi-circularly curved; sloping laterally from the elevated mid-region and then gradually becomes flatly oval posteriorly. However, caudo-ventro-laterally, the ventral surface of the free portion of the tongue is depressed on either sides of the median line. The depression marks the site of origin of the muscle hypoglossus anterior and encloses the rostral parts of the paired ceratobranchiale (Fig.1b).

Meanwhile, in the common hoopoe, the free portion of the tongue (FRT) is flat, with rounded rostral tip and broad caudal margin. Its postero-dorsal surface bears lingual tubercles which varied in number in the investigated specimens, from three pairs in some specimens to many of varying sizes in others (Figs.14a,b&21a).

The wide caudal margin of the free portion of the tongue bears a pair of swollen lingual wings (LW) that ends with paired large postero-lateral papillae one on each side. The large postero-lateral papilla is short, conical, puffed rostrally and pointed near the base. It bifids into small external papilla and large internal one as well as, it is converged toward the mid-dorsal line. In some specimens there is an additional pair of papillae that projects one on either side of the middle of the free portion (Figs.14b&22a). Moreover, there are 3 short lingual papillae of varying sizes which are arranged in single row on either sides of the caudal margin of the free portion between the two large postero-lateral papillae toward the laryngeal area (Papillae lingualis, PL) (Figs.14b). In both bird species, the free portion of the tongue connects with the floor of the mouth through extension of the ventral overspreading skin of the floor of the mouth is known as frenulum which is a fleshy and non-pigmented triangular-shaped in the laughing dove (Fig.1b), while broad in the common hoopoe (Fig.24). Posterior to the free portion, there is

laryngeal area which is distinguished into two regions; the rostral preglottal area (PGR) and the caudal laryngeal mound (LRM). In the laughing dove, the laryngeal area is approximately equal in length to the free portion of the tongue (12-14 mm), but it is wider than it (Figs.1a& 9). Meanwhile, in the common hoopoe the length of the laryngeal area is approximately 9 mm, where it represents twice the length of the free portion of the tongue (Figs.14a).

In the laughing dove, the preglottal area is short triangular to oval-shaped. It is demarcated interiorly by the caudal margin of the free portion and posteriorly by the glottis and it is supported by the hyoid skeleton. Many orifices represent the secretory pores of the glandula preglottalis (GL.PRG) which open on the dorsal surface of the preglottal area (Figs.1a&9). The laryngeal mound (LRM) lies caudal to the preglottal area where, it incubates the wide glottis at its mid-dorsal line. Also, the laryngeal mound is supported by the laryngeal skeleton. That laryngeal mound bears 6-7 thick and short conical papillae (laryngeal papillae, LRP) of varying sizes. These laryngeal papillae are arranged irregularly on either side of the mid-dorsal line. Moreover, there are about 10 conical and pointed laryngeal papillae (LRP) of different sizes which are arranged in single row on either side of the caudal margin of the laryngeal mound. Also, the secretory pores of the glandula laryngealis (GL.LR) open on the dorsal surface of the laryngeal mound (Figs.1a&9). In the common hoopoe, the preglottal area is short and oval-shaped, that is situated between the posterior margin of the free portion anteriorly and glottis posteriorly. It bears on its dorsal surface beneath the covering epithelium, the preglottal gland (GL.PRG) (Figs.14a,b&23a,b).

Posterior to the preglottal area, occurs the laryngeal mound (LRM) that incubates the wide glottis at its mid-dorsal line as well as the laryngeal gland which is distributed beneath its dorsal surface (GL.LR). The mid-dorsal base of the laryngeal mound caudal to the glottis bears 2 pairs of thick conical papillae of different sizes. Also, from 9 to 10 conical laryngeal papillae of different sizes are arranged in two rows on either side of the base of the caudal area of the laryngeal mound (Figs.14a&25a).

THE LINGUAL EPITHELIUM

The lingual epithelium of both bird species is covered by stratified squamous epithelium. The light microscopic investigation of the lingual epithelium of the laughing dove revealed that the dorsal epithelium (DE) covering the free portion of the tongue is thicker than that of the ventral one (VE) (Figs.2a,b&3). The epithelium of the most anterior-third of the free portion is furnished dorsally by detached keratin, with the appearance of the deciduous epithelium outside to the keratinized layer which is thin and their cells are filled with keratin granules (Fig.2a,b).

Histological investigation along the whole length of the free portion of the tongue of the laughing dove revealed that the mucosa of the dorsal epithelium is characterized by the presence of downward folds (Reteridges) which interdigitate with the upper projections of the submucosa (Dermal papillae, DP) resulting in the formation of the mucoso-submucosal junctions (Fig.4).

The long paired lingual wing (LW) and their lingual papillae (LP) are enveloped by a non-keratinized stratified squamous epithelium interspersed with scattered soft keratinized cells. Each lingual papilla is composed of the mucosa and submucosa (SM). The submucosa gives rise to the core of the papillae, while the mucosa, which is non-keratinized epithelium, forms the outer envelope of the papillae. The envelope of the lingual papillae has short dermal papillae (Figs.4a,b&10a,b).

SEM investigation of the dorsal epithelium of the anterior third of the free portion of the tongue of the laughing dove, showed the presence of filiform papillae emerging from the deciduous epithelium that is directed medially towards the median sulcus (Sulcus Lingualis, SL). By using high magnification, the investigation showed presence of microridges on the surfaces of the filiform papillae (Fig.8a,b). However, the keratinization decreases posteriorly of the epithelium covering the paired lingual wing and their lingual papillae to become smooth with scattered soft keratinized cells (Fig.10a,b). The ventral epithelium (VE) covering the free portion of the tongue of the laughing dove is highly keratinized stratified epithelium forming the differentiated lingual nail (LN). That epithelium is thinner than that of the dorsal one and is characterized by the presence of many short dermal papillae (Fig.2a,b). Therefore, the ventral epithelium covering the free portion of the tongue exhibits a gradual decreasing of keratinization until it becomes non-keratinized epithelium at the end of the anterior third of the ventral surface of the free portion (Fig.4a).

The dorsal surface of the preglottal area of the laughing dove (PGR) is furnished by non-

keratinized squamous epithelium with flattened nuclei which is characterized by the absence of true keratin. The preglottal epithelium is characterized by the appearance of the dermal papillae (DP) which are shorter and fewer than that of the dorsal epithelium of the free portion of the tongue (Fig.8c). SEM investigation reveals that the epithelium covering the preglottal area is smooth with detached keratin with the presence of intercellular borders of the outer cellular layer and the pores of the glandula preglottale (GL.PRG) (Fig.11a,b).

The laryngeal mound of the laughing dove (LRM) is covered dorsally by non-keratinized squamous epithelium without dermal papillae (Fig.5). SEM investigation along the whole length of the laryngeal mound revealed that the dorsal surface of the laryngeal mound is smooth with detached keratin and many pores of the glandula laryngealis (GL.LR) (Fig.12a,b).

The covering epithelium of the frenulum (F) and the paralingual region (PL) is transitional-like one. This transitional-like epithelium is somewhat similar to the squamous epithelium when it is stretched, but when it is collapsed the nuclei of the more superficial cells becomes rounded. Moreover, the covering epithelium of the floor of mouth at the connection with the tongue (Frenulum and paralingual) is characterized by the presence of multiple dermal papillae (DP) (Fig.6). SEM investigation revealed the presence of the mucosal ridges covered by detached epithelium. This type of epithelial cells which has the ability to stretch and collapse, give the free portion of the tongue the ability to protract and retract (Fig.13).

Meanwhile, in the common hoopoe, *Upupa epops*, the light microscopic investigation of the lingual epithelium revealed that the dorsal epithelium (DE) of the free portion of the tongue is relatively thicker than that of the ventral one (VE) (Fig.15a). Both the dorsal and ventral epithelium covering the whole surface of the free portion of the tongue is furnished by α keratin that shown well in scanning electron microscopy (SEM) investigation, while disappears during routine histological preparations (Figs.21a&24). That epithelium is furnished with scattered filiform papillae with microridges and the borders of the deciduous cells are well observed (Figs.21b). Meanwhile, the epithelium covering the lingual tubercles is furnished dorsally by true keratin Figs.15b&16). The long paired lingual wing (LW) is enveloped by a true keratinized stratified squamous epithelium without dermal papillae (DP). Meanwhile, the surface of the lingual papillae (LP) is furnished by keratinized epithelium with short dermal papillae. Each lingual papilla is composed of mucosa and submucosa (SM). The submucosa gives rise to the core of the papillae, while the mucosa forms the outer envelope of the papillae with long dermal papillae. The microridges and the borders of the deciduous cells are well observed by SEM investigation (Figs.15b,16&22a,b).

The preglottal area (PGR) is furnished by non-keratinized squamous epithelium with the appearance of short dermal papillae (DP). The epithelium of the preglottal area is thinner than that of the dorsal and the ventral epithelium covering the free portion of the tongue (Fig.17). SEM investigation shows the presence of detached keratin with microridges and intercellular borders of the outer cellular layer, as well as the pores of the glandula preglottale (GL.PRG) which are surrounded by dense mucous secretion (Figs.23a,b).

The laryngeal mound (LRM) is covered dorsally by non-keratinized squamous epithelium with few scattered and short laryngeal dermal papillae. The surface of these laryngeal papillae is covered by true keratinized squamous epithelium (LRP) (Fig.18). The histological structure of these papillae resembles that of the lingual ones. SEM investigation along the whole length of the laryngeal mound revealed that the dorsal surface of the laryngeal mound is smooth with detached keratin. The microridges and intercellular borders of the outer cellular layer, as well as many pores of the laryngeal gland (Glandula laryngealis, GL.LR) are well observed (Fig.25a,b). The covering epithelium of the frenulum (F) is non- keratinized stratified squamous epithelium with flattened nuclei in the outer cellular layers with the presence of multiple dermal papillae (DP) (Fig.19).

THE SALIVARY GLANDS

The salivary glands of both bird species are classified according to their location; the laughing dove possess the glandula Sublingualis (GL.SL),glandula Preglottalis (GL.PRG), glandula laryngealis (GL.LR), and glandula lingualis (GL.L). The latter one in the common hoopoe is not evident.

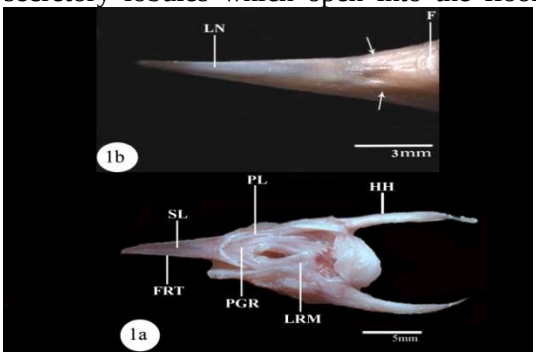
In the laughing dove, the glandula lingualis is a paired one which extends on the posterior half of the dorso-lateral surface of the Os paraglossale (PG) and their posterior processes (P.post.pg) (Fig.4). Meanwhile, the glandula lingualis is a compound tubulo-alveolar type , each gland consists of two to

three lobules and is encased inside a flexible collagenous connective tissue sheath. The glandular lobules produce a large amount of mucopolysaccharides which exhibit a positive PAS reaction (Fig.7a,b). The glandula lingualis performs many functions; it lubricates the food items by its mucoid secretion. Meanwhile, the gland acts as a hydroskeleton due to its relationship with the Os paraglossale. Moreover, the gland which acts as a hydraulic structure contributes in the transformation of the dorsal surface of the free portion of the tongue which is covering by a parakeratinized epithelium.

The preglottal gland in both bird species is unpaired one which occupies the preglottal area (PGR) between the paired lingual wing of the free portion of the tongue and rostral to the glottis. The preglottal gland is a compound tubulo-alvealor that consists of many secretory lobules which produce a large amount of mucopolysaccharides and exhibit a positive PAS reaction. The glandula preglottalis opens at the dorsal epithelial surface by multiple openings which deliver their mucoid secretion for lubricating the food items and facilitate their passage (Fig.7c,d). Meanwhile, the glandula preglottalis acts as a hydroskeleton due to its relationships with the lingual papillae, the glottis, and the lingual muscles which located in the preglottal area.

The glandula sublingualis of both bird species is paired gland that is embedded in the skin and extends along the lateral sides of the floor of the mouth, dorsal to the posterior two thirds of the mylohyoideus muscle. The glandula sublingualis is a compound tubulo-alveolar type that consists of many secretory lobules which open into the floor of the mouth via many orifices. The gland is enveloped by glandular capsule which adheres medially with the lateral borders of the epimysium of the posterior portion of the genioglossus muscle and laterally with the anterior portion of the branchiomandibularis muscle. The sublingual gland exhibits a positive PAS reaction which means that it secretes mucopolysaccharides (Figs.7e,f&20c,d). That mucous secretion lubricates the food items during its passage in the buccal cavity, as well as, it acts as a hydroskeletal structure due to its connection with the genioglossus and the branchiomandibularis muscles.

The glandula laryngealis of both bird species is a paired gland which exhibits a triangular shaped in the laugh dove embedding within the skin of the paired laryngeal wing caudal the laryngeohyoideus muslce (M.lrh) and has paired anterior extensions that extend rostrally parallel to the posterior portion of the glottis. This gland of the laugh dove is a compound tubulo-alveolar type that consists of many secretory lobules which open into the dorsal epithelial surface by multiple openings and deliver their mucoid secretion for lubricating the food items and facilitate their passage (Figs.7g). While in the common hoopoe, the glandula laryngealis is formed of multiple lobules of simple alveolar acini embedded in the skin of the laryngeal wings. The glandula laryngealis of both bird species exhibits highly positive PAS reaction which means that it secretes a large amount of mucopolysaccharides (Fig.20f). Another role of the glangula laryngealis, that acts as a hydroskeletal structure due to its connection with the cricoarytenoideus and the laryngeohyoideus (M.lrh) muscles.



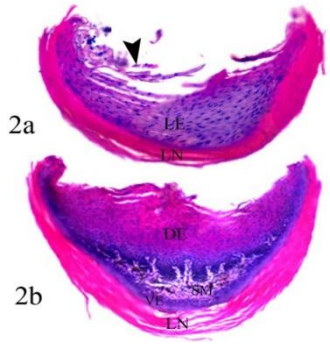
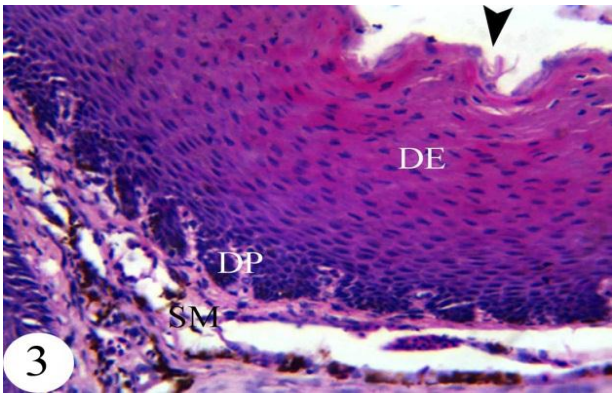
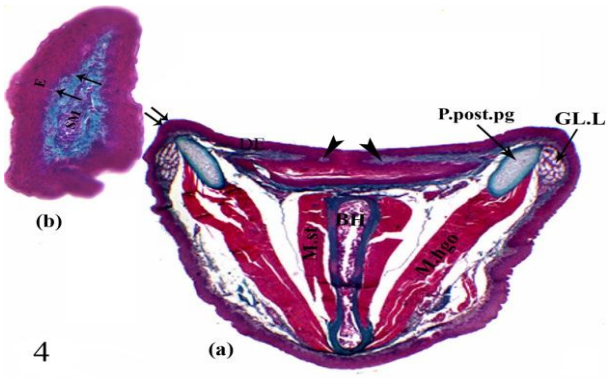
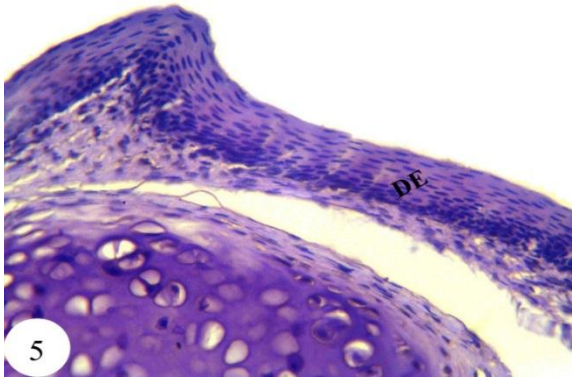


Fig.1a. Photomicrograph of dorsal surface of the tongue of the laughing dove, showing the free portion (FRT), preglottal region (PGR), laryngeal mound (LRM), hyoid horns (HH), the paralingual (PL) and Sulcus Lingualis (SL). b. Ventral surface showing the lingual nail (LN) and frenulum (F). The lateral margins of the frenulum enclose the rostral parts of the paired ceratobranchiale. **Fig.** 2a. Transverse section through the anterior tip of the free portion of the tongue of the laughing dove, showing the lingual epithelium (LE) is furnished by non-true keratin "parakeratosis" with some projections on the surface beside their cells are filled by keratin granules (arrowhead). In addition, the lingual nail (LN) (H&E, X200). b. Transverse section through the most anterior-third of the free portion, showing the dorsal epithelium (DE) (with well-developed dermal papillae) is thicker than the ventral epithelium (VE), in addition, the lingual nail (LN) and submucosa (SM) (H&E, X100).







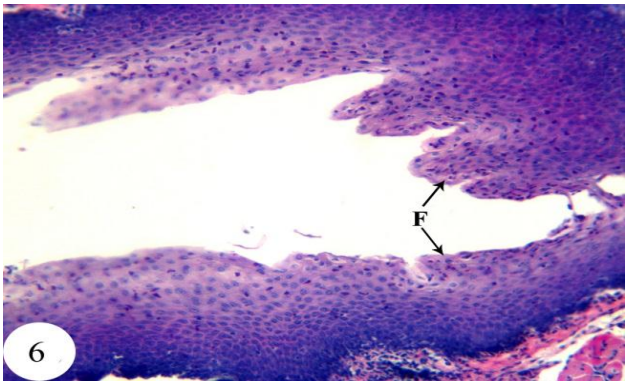
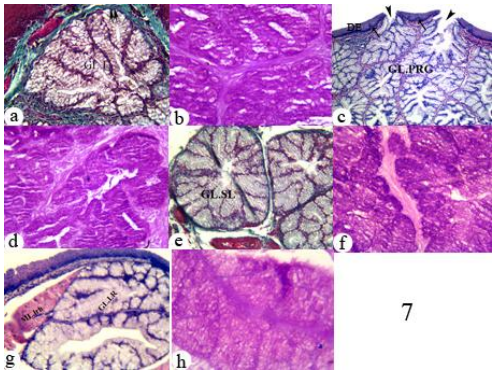


Fig. 3. Transverse section through the most the anterior-third of the free portion of the tongue of the laughing dove, showing the dorsal stratified squamous epithelium (DE), Sulcus Lingualis (arrowhead), and the short dermal papillae (DP), in addition, the submucosa (SM) (H&E, X400).

Fig.4. a. Transverse section through the posterior end of the free portion of the tongue of the laughing dove, showing the lingual wing with its supporting element, the posterior process of paraglossale (P.post.pg), and the lingual papillae (LP). In addition, the Glandula Lingualis (GL.L), the muscle hypoglossus obliquus (M.hgo), and the muscle stylohyoideus (M.st). b. the structure of the lingual papilla (LP), the submucosa forms the core of the papillae, while the non-keratinized mucosa, forms the outer envelope of the papillae. In addition, the short dermal papillae (arrow) (Masson's trichrome stain, X32).

Fig.5. Transverse section through the laryngeal mound of the tongue of the laughing dove, at the end of the glottis showing, the non keratinized squamous epithelium covered the laryngeal mound without dermal papillae (H&E, X200).

Fig.6. Transverse section through the frenulum (F) that is formed of transitional-like epithelium, and has the ability of stretching and collapsing (H&E, X100).



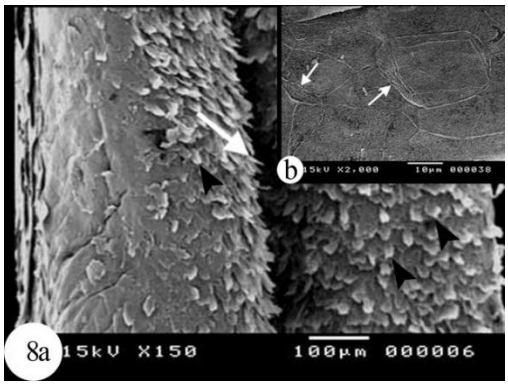


Fig.7. Transverse section of the salivary glands of the laughing dove (a,b. Masson's trichrome stain, X400&PAS, X400) Glandula Lingualis (GL.L), (c,d. H&E, X200& PAS, X400) glandula preglottalis (GL.PRG), (e,f. Masson's trichrome stain, X400&PAS, X400) Glandula Sublingualis (GL.SL) and (g,h. H&E, X200&PAS, X400) glandula laryngealis (GL.LR).

Fig.8. a. Scanning electromicrograph of the dorsal surface of the Anterior-third of the free portion of the tongue of the laughing dove, showing the presence of filiform papillae are directed medially towards the Sulcus Lingualis (arrow) (X150). b. High magnification, showing the presence of microridges on the surfaces of the filiform papillae (arrow) (X2000).

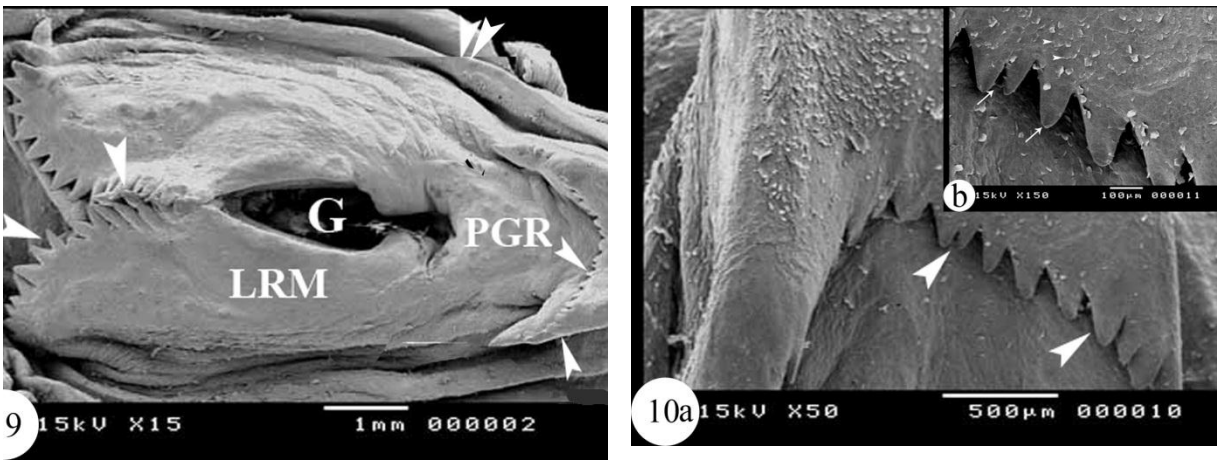


Fig.9. Scanning electromicrograph along the dorsal surface of the tongue of the laughing dove, showing the lingual wing with one row of lingual papillae ((arrowhead)), the triangular to oval-shaped preglottal area (PGR) that demarcated anteriorly by the caudal margin of the free portion of the tongue, and posteriorly by the glottis (G), the laryngeal mound (LRM) caudal to the preglottal area with the laryngeal papillae arranged in one row on its caudal margin (arrow). In addition, a shallow groove where it abuts the paired ceratobranchiale (double arrowhead) (X15).

Fig.10. a. Scanning electromicrograph of the dorsal surface of the lingual wing of the laughing dove, showing one row of the backward lingual papillae (arrowhead) (X50). b. High magnification, showing decreases of keratinization of the epithelium covering the paired lingual wing (LW) and their lingual papillae (LP) with scattered soft keratinized cells (arrow) (X150).

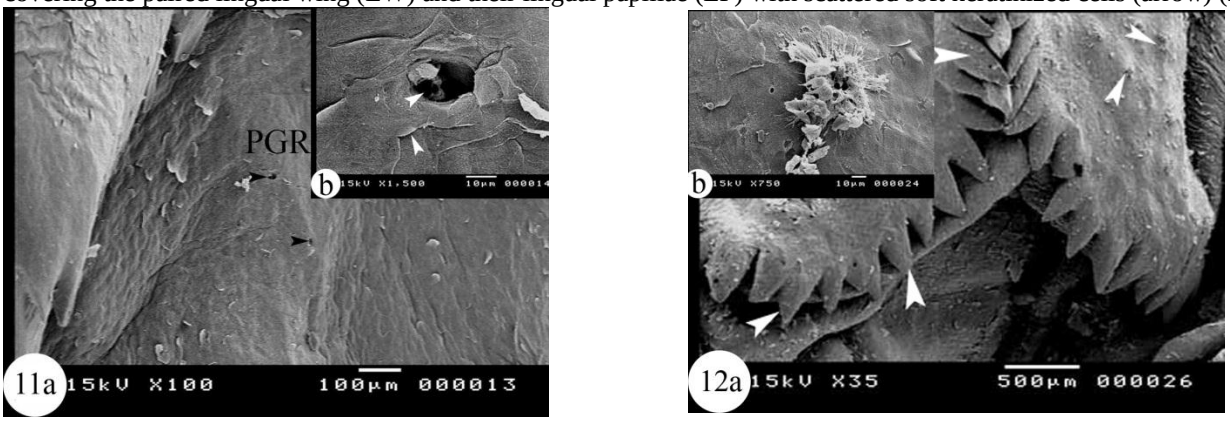
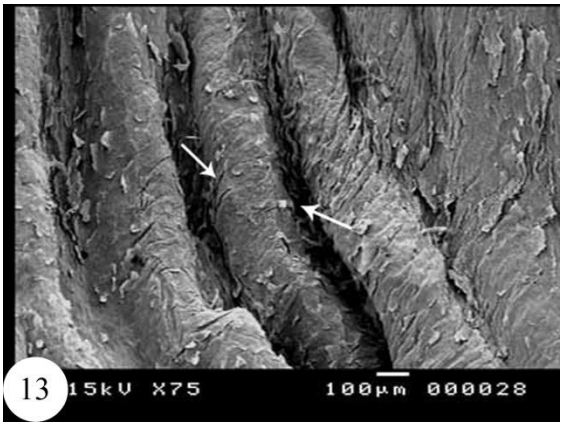


Fig.11. a. Scanning electromicrograph of the dorsal surface of the preglottal region of the tongue of the laughing dove, showing, the epithelium covering the preglottal area is smooth with detached keratin. In addition, the appearance of the orifices of the glandula preglottalis (arrowhead) (X100). b. High magnification, showing the presence of the intercellular borders of the outer cellular layer of the epithelium covering the preglottal area and an orifice of the glandula preglottalis (arrowhead) (X500).

Fig.12. a. Scanning electromicrograph of the caudal margin of the laryngeal mound (LRM) of the tongue of the laughing dove, showing, the dorsal surface of the laryngeal mound is smooth with detached keratin and many pores of the glandula laryngealis (GL.LR) (arrowhead) (X35). b. High magnification, showing the epithelium covering the laryngeal mound with detached keratin and a pore of the glandula laryngealis is surrounded by mucous secretion (X750).



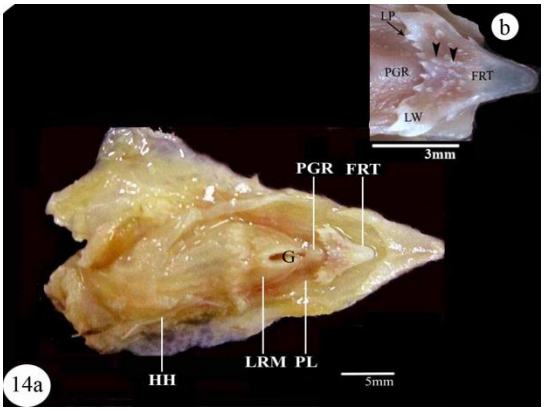
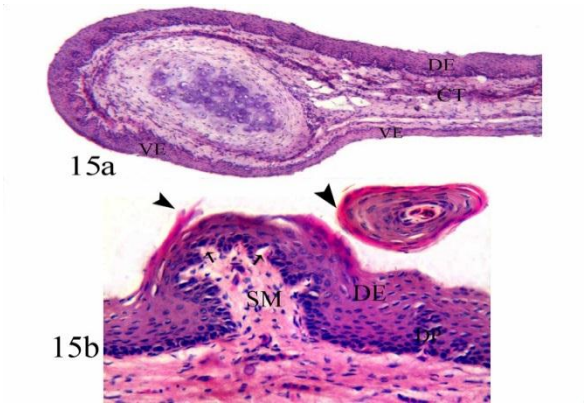


Fig.13. Scanning electromicrograph of the dorsal surface of the paralingual in the laryngeal area of the tongue of the laughing dove, showing the wrinkled epithelium that forms the mucosal ridges with detached keratin (arrow) (X75).

Fig.14. a. Photomicrograph of the dorsal surface of the tongue of the common hoopoe, showing the free portion (FRT), preglottal region (PGR), laryngeal mound (LRM), and hyoid horns (HH), in addition, the paralingual (PL). b. High magnification, showing the lingual tubercles (arrowhead) scattered on the free portion of the tongue (FRT), one row of lingual papillae (LP) on the caudal margin of the paired lingual wing. In addition, the preglottal region (PGR) and the laryngeal mound (LRM).



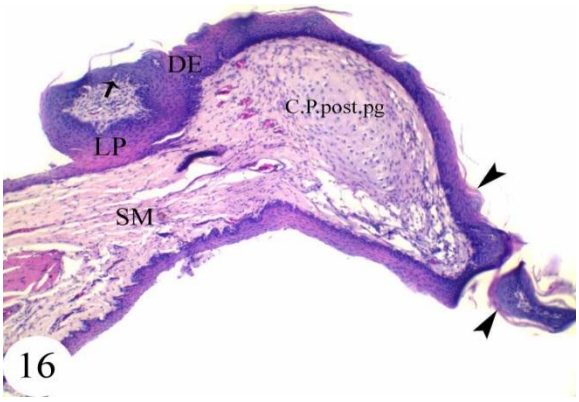


Fig.15. a. Transverse section through the right half of the most anterior-third of the free portion of the tongue of the common hoopoe, showing the dorsal (DE) and ventral epithelium (VE) (H&E, 100x). b. High magnification, showing the dorsal epithelium (DE), the dermal papillae (DP) and a lingual tubercle (arrowhead) that covers by a layer of true keratin, in addition the submucosa (SM) (H&E, X400).

Fig.16. Transverse section through the lingual wing of the free portion of the tongue of the common hoopoe, lingual papillae (LP) which are covered by a true keratinized epithelium (arrowhead), and have dermal papillae (arrow), in addition the cartilaginous tip of the processus posterior paraglossale (C.P.post.pg) (H&E, X 100).

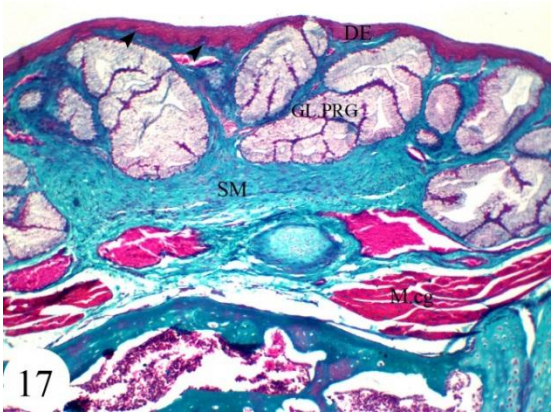
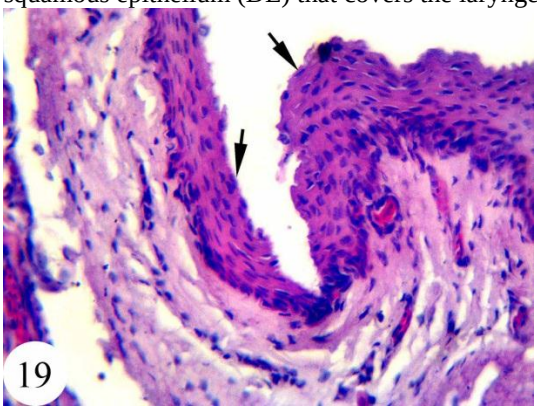


Fig.17. Transverse section through the anterior portion of the preglottal area of the tongue of the common hoopoe, showing the non-keratinized dorsal epithelium (DE) covers the preglottal area with short dermal papillae (arrowhead), in addition, the preglottal gland (GL.PRG), the submucosa (SM), and the muscle ceratoglossus (M.cg) (Masson's trichrome stain, X100).

Fig.18. Transverse section through the middle region of the laryngeal mound of the common hoopoe, showing the keratinized squamous epithelium (DE) that covers the laryngeal mound and the laryngeal papillae (arrowhead) (H&E, X100).



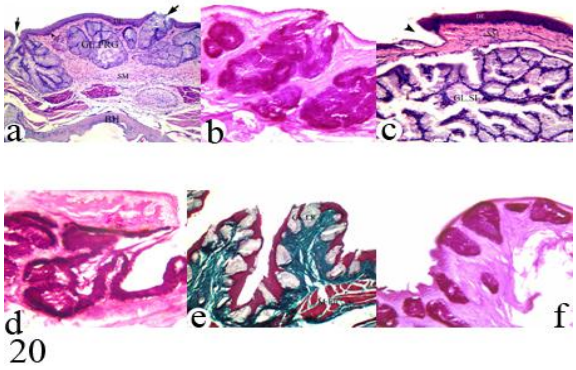
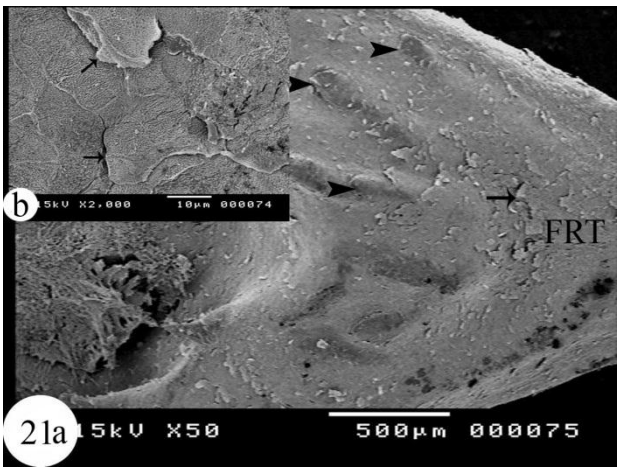


Fig.19. Transverse section through the frenulum (F) which consists of non-transitional stratified squamous epithelium with the outer cellular layers of flattened nuclei (arrow) (H&E, X400).

Fig.20. Transverse section of the salivary glands of the common hoopoe (a,b. H&E, X32 & PAS, X200) glandula preglottalis (GL.PRG), (c,d. H&E, X200& PAS, X400) Glandula Sublingualis (GL.SL) and (e,f. Masson's trichrome stain, X100& PAS, X200) glandula laryngealis (GL.LR).



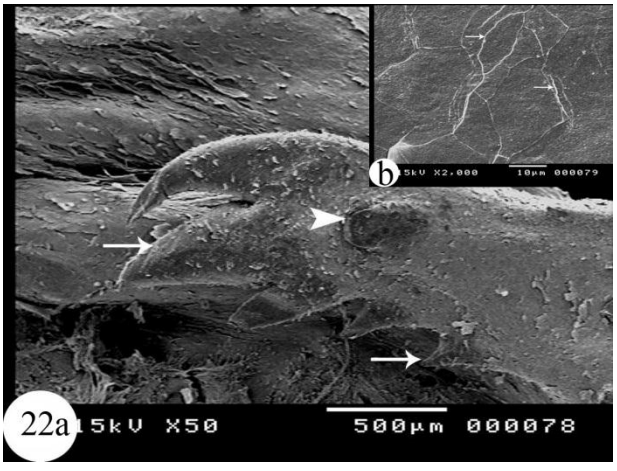
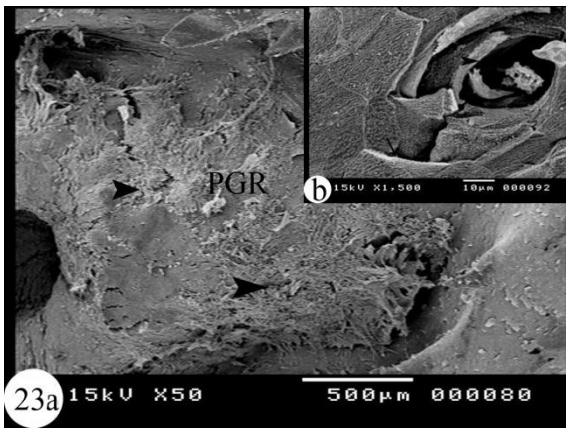


Fig.21. Scanning electromicrograph of the dorsal surface of the posterior part of the free portion of the tongue (FRT) of the common hoopoe, showing the scattered lingual tubercles (arrowhead) (X50). b. High magnification, showing the microridges and borders of the deciduous epithelium (arrow) (X2000). Fig.22. Scanning electromicrograph of the dorsal surface of the

lingual wing of the free portion of the tongue of the common hoopoe, showing the deciduous epithelia cover the lingual papillae (arrow) and the lingual tubercles (arrowhead) which are furnished by true keratin (X50). b. High magnification, showing the microridges and borders of the deciduous epithelium (arrow) (X2000).



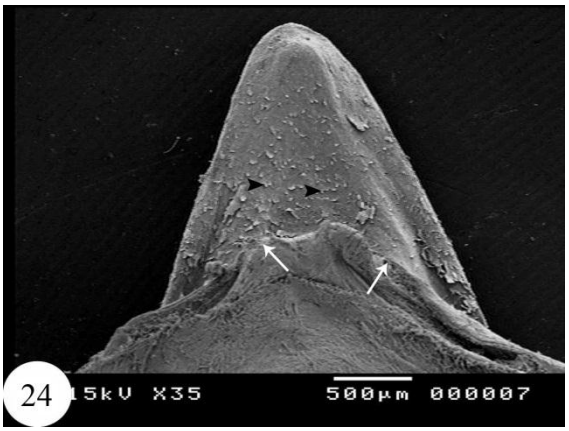


Fig.23. Scanning electromicrograph of the dorsal surface of the preglottal region of the tongue of the common hoopoe, showing the triangular to oval-shaped preglottal area (PGR) that furnished by detached keratin (arrowhead) (X50). b. High magnification, showing the presence of microridges with intercellular borders of the outer cellular layer of the epithelium covering the preglottal area, as well as the pore of the glandula preglottalis (arrowhead) (X500).

Fig.24. Scanning electromicrograph of the ventral surface of the anterior part of the free portion of the tongue of the common hoopoe, showing the deciduous epithelium, which is furnished by α keratin (arrowhead), in addition the wide frenulum (X100).

Fig.25. Scanning electromicrograph of the laryngeal area of the tongue of the common hoopoe, showing the laryngeal mound (arrowhead) caudal to the preglottal area with the laryngeal papillae arranged in two rows on the caudal margin of the laryngeal mound (double arrows) (X15). b. High magnification, showing the epithelium covering the laryngeal mound with detached keratin and microridges of the outer cellular layer (arrow), as well as a pore of the glandula laryngealis (arrowhead) (X1500).

DISCUSSION

The tongue of the laughing dove, *Streptopelia senegalensis* and the common hoopoe, *Upupa epops* like almost avian tongue is a complex biomechanical organ, which is connected with the floor of the mouth. Stratification and keratinization of the avian lingual epithelium is a common feature[(10)]**.** Also, non-keratinized and parakeratinized areas were found among the avian tongue.

The present results indicated the modifications of the epithelium covering the whole length of the tongue of the two studied bird species.

These modifications are specific to each species. The epithelium covering the tongue of the laughing dove exhibits extreme regional variations. The dorsal surface of the anterior part of the free portion of the tongue is covered by a parakeratinized squamous epithelium with the appearance of deciduous cells outside to the keratinized layer. However, the keratinization decreases posteriorly at the paired lingual wing. The highly thick layer of true keratin is found in the lingual nail underlies the ventral epithelium of the anterior third of the free portion of the tongue which supported by the

cartilaginous Os paraglossale. The thick layer of true keratin decreases gradually until it disappears at the end of the anterior third of the free portion.

Occurrence of more stratification in the tongue of the laughing dove may counteract the abrasive forces related to the hard texture of the grains with hard seeds which they take in the form of food.

The investigation of the lingual nail during the movement of the tongue illustrates that; it is bendable. Thus it can change its shape according to its use. Therefore, it is affected by the internal forces acting on the tip of the tongue. Although, the lingual nail is a supportive structure for the anterior half of the free portion of the tongue, it is acting as a spoon with its pointed anterior edge hence facilitating picking up the food items of seeds and grains, as well as, it also help the bird in drinking. Moreover, the lingual nail supports the anterior third of the free portion of the tongue during its bending by the contraction of the muscle hypoglossus anterior.

However, the tongue of the common hoopoe, exhibits some specific histological modifications. The stratification of the epithelial layer along the whole length of the tongue is less pronounced than that of the laughing dove and it may due to the less hard diet intake i.e. worms; these results agree with those of **Singh**[11]**.** Both the dorsal and ventral surfaces of the free portion of the tongue are furnished by non-keratinized squamous epithelium except that constitutes the lingual wings, lingual tubercles and lingual papillae which are furnished by true keratin. This type of keratins appears to be produced only in the differentiated epithelial cells of the antero-ventral region of the tongue (the lingual nail)[12]**. Shawki** [13] concluded that the highly keratinized epithelium establishes the most prominent keratinized elements among the avian tongue; the lingual nail, papillae, the laryngeal papillae and the laryngeal mound.

Biomechanical analysis has demonstrated that α keratins are produced in essentially all vertebrate epithelium[14]**,** but the β keratin are detected only in specific epithelial tissues of birds and reptiles[15]**.** Also, non-keratinized and parakeratinized epithelia were found among the avian tongue. **Carver and Sawyer**[12] concluded that in parrot, the β keratin is expressed in the antero-ventral region of the free portion of the tongue, while α keratin is detected in all epithelium of the tongue.

Moreover, the presence of the lingual tubercles distributed along the caudal region of the dorsal surface of the free portion of the tongue of the common hoopoe may help in bushing the food items internally. The presence of these tubercles meeting the palatine papillae on the roof of the mouth (palate) [16] and hence perform bushing of food well into the pharynx. Meanwhile, the present authors suggest that the variation in number of the lingual tubercles of the common hoopoe, may point out divergence of several subspecies.

The parakeratinized epithelium of the tongue of both the laughing dove and the common hoopoe is emerging with filiform papillae containing microridges. These microridges may represent an adaptation of the epithelial cells of the tongue of the laughing dove for providing friction or may play a role in holding and spreading mucus in the case of the common hoopoe. **Fahrenbach and Knutson**[17] studied surface adaptation of the vertebrate epidermis to friction, and suggested that microridges play a role in providing friction. Also, **Shawki and Ismail**[18] supposed the same role of the microridges in the case of the common moorhen and were suggested that the spreading of mucus is not necessary for this aquatic feeder bird. **Sperry and Wassersug** [19] suggested that microridges might play a role in holding and spreading mucus. The same result was suggested by **Shawki** [13] in the tongue of the common kestrel like that mentioned on the kite which is also a predator bird living in dry condition.

The occurrence of the above mentioned type of stratified epithelium is common in birds e.g. in the chicken tongue[20], terminal modification of the dorsal epithelium is more distinctly different in the anterior and posterior regions, so-called " filiform papillae" or distinct protrusions of the deciduous epithelial cells that are widely distributed over the anterior region of the chick tongue, whereas they are not present in the posterior region. In the white-tailed eagle[21], most of the dorsal surface of the lingual corpus and root is covered with the intensively desquamate non-keratinized epithelium "parakeratinized epithelium" and suggested that it may be considered species specific trait.

In this study, parakeratinized epithelium of transitional-like type with rounded nuclei lines the connection of the tongue with the floor of the mouth (frenulum) of the laughing dove. The main hyoid joints region, at which arise the main movements of

the tongue. That transitional epithelium has a stretching ability and consequently its existence is a demand adaptive for the mechanical performance of the tongue during its protraction and retraction and during the elevation and depression of the free portion of the tongue. Meanwhile, in the common hoopoe, this region is less wrinkled and is covered by non-transitional stratified squamous epithelium with flattened nuclei, which means that this epithelium lack the ability for stretching, and therefore restricts the mechanical performance of the tongue. The lingual and laryngeal papillae of the laughing dove are smooth, pointed and posteriorly directed with detached keratin. While in the common hoopoe, the lingual and laryngeal papillae are covered by a true keratinized squamous epithelium. It has been expected that the lingual and laryngeal papillae may have a role in processing the food items. As regards the number and distribution of lingual papillae and laryngeal papillae, **Bhattacharyya**[(22)] reported that it cannot be clearly ascertained as to how far these structures are phylogenetic and how far adaptive. Salivary glands as secretory organs always attract the attention of the physiologists and histologists. However, from the functional anatomical point of view the authors of the present work classified the salivary glands as hydrostatic structures which transferred from soft into rigid ones, and then they may be considered as skeletal-like elements. The number and size of the salivary glands in the lingual apparatus of the laughing dove is more and larger than that of the common hoopoe due to the reduction of its tongue. The lingual apparatus of the laughing dove has four salivary glands of compound type, while the lingual apparatus of the common hoopoe has only three salivary glands with absence of the lingual on.

Each salivary gland of the two studied species is encased by envelop which consists of collagenous connective tissue. That tissue may bend, but not stretch. [(23)] indicated that the secretion of these lingual glands was collected in the sub epithelial chamber with the wide orifices, and then was effectively evacuated to the surface of the tongue. The liquid content of these glands is mucopolysaccharides (mucin). **Homberger**[(24)] stated that the mucous is visco-elastic element. So, the salivary glands interact with the mechanical performance of the tongue as hydraulic structures depending on the physical properties of their structures, as well as, the relationships of the glands with the other structural components of the tongue. **Garqiulo *et al***[(23)] stated that one of the main components of the secretion of the salivary glands is the glutinous mucus which might act as inhibitors of some bacterial enzymes. In addition, the salivary glands may act as cushion between the contracting muscles, for examples; the glandula sublingualis and mandibularis. Also, the salivary glands act as hydroskeletal elements, which are represented by the glandula sublingualis, preglottalis and laryngealis.

Meanwhile, the shape of the salivary glands as hydraulic structure and the amount of its pressure (turgidity) can be varied by the application of external forces on the fluid container or by changes in the fluid volume[(25)]. Meanwhile, the glandula lingualis of the laughing dove may plays essential and complicated role during the movement of the free portion and the anterior tip of the tongue, as well as, in adjusting the upright position of the lingual papillae during the food passage. In addition to the secretory and hydrostatic functions of these glands, they may act as skeletal elements since they are enveloped by connective tissue which serves as an insertion site for some lingual muscles. Absence of the lingual gland of the common hoopoe may due to the great reduction of the free portion of the tongue and the soft and moist food kind intake.

In conclusion, the keratinized epithelium covering the dorsal surface and forms the sulcus lingualis and the lingual nail covering the ventral surface of the free portion of the tongue of the laughing dove as well as the transitional epithelium covering the frenulum construct to be bendable (up down movement) make the free portion like spoon and helps the bird for drinking and packing seeds up with the aid of the lingual gland and the paraglossus process. Meanwhile, the epithelium covering the free portion and frenulum of the common hoopoe exhibits less mobility in addition to absence of the lingual gland hence the bird depends in its first action of feeding on the movement of the jaws and beak. On the other hand, the dorsal epithelium is provided by tubercles aids in pushing the food items toward the laryngeal area.

The epithelium covering the laryngeal area of the two bird species with its derivatives (preglottal and laryngeal glands and dermal papillae) are modified for moistening and gliding the food items toward the esophagus.

REFERENCES

1. **Roth G and Wake D B (1989):** Conservatism and innovation in the evolution of feeding in vertebrates. In: Complex Organismal Functions: Integration and Evolution in Vertebrates. (Wake, D.B., Roth, G. (eds). John Wiley and Sons New York, pp: 7–21.
2. **McLelland J (1979):** Digestive system. In: Form and Function in Birds. King, A.S. and McLelland, J. (eds). Academic Press. London, pp**:** 69-181.
3. **Homberger D G and Meyers R A (1989):** Morphology of the lingual apparatus of the domestic chicken, *Gallus gallus*, with special attention to the structure of the Fasciae. The American Journal of Anatomy, 186**:** 217-257.
4. **Igwebuike U M and Anagor T A (2013):** The morphology of the oropharynx and tongue of the muscovy duck (*Cairina moschata*). Vet. Arhiv., 83: 685-693.
5. **Trivedi V and Soni V C (2013):** Epidermal structures of the bill and lingual of the eurasian collared dove (*Streptopelia decaocto*). Journal on New Biological Reports, 2(1): 44-54.
6. **Erdogan S and Perez W (2015):** Anatomical and scanning electron microscopic characteristics of the oropharyngeal cavity (Tongue, Palate and Laryngeal Entrance) in the Southern Lapwing (Charadriidae: *Vanellus chilensis*, Molina 1782): Acta Zoologica (Stockholm), 96: 264–272.
7. **Shawki N A Abdeen A M and Mahmoud F A (2016):** Functional morphological study of the oropharyngeal cavity and the tongue of the common kestrel, (*Falco tinnunuculus*).The roof of the oropharyngeal cavity. IJRSZ., 1(3): 16- 26.
8. **Huang R, Zhi Q ,Izpisua-Belmonte J C, Christ B and Patel K (1999):** Origin and development of the avian tongue muscles. Anat. Embryol., 200**:** 137-152.
9. **Drury RA and Wallington EA (1980):** Carleton's Histological Techniquies. 4th ed. Oxford University Press. London, NewYork, Toronto, pp: 214-215.
10. **Kobayashi K Kumakura M Yoshimura K Inatomi M and Asami T (1998):** Fine structure of the tongue and lingual papillae of the penguin. Arch. Histol. Cytol., 61**:** 37–46.
11. **Singh SP (1974):** Morpho-histological observations on the tongues in some birds namely *Ploceus philippinus*, *Upupa epops*, *Megalaima haemacephala* and *Cinnyris asiatica*. Zoo1. Surv. India, Kolkata, 86: 75-84.
12. **Carver WE and Sawyer RH (1989):** Immunocytochemical localization and biochemical analysis of α and β keratins in the avian lingual epithelium.Am. J. Anat., 184**:** 66–75.
13. **Shawki N A (2016):** Jaw Apparatus, Kestrel and Budgerigar. 1st., Lambert Academic Publishing. Germany.
14. **Sun T T Shih C and Green H (1979):** Keratin cytoskeletons in epithelial cells of internal organs. Proc. Natl. Acad. Sci. USA., 76**:** 2813-2817.
15. **Landman L (1986):** The skin of reptiles**:** epidermis and dermis. In: Vertebrates Biology of the Integument. Bereiter-Hahn, J., Matoltsy, A.G. and Richards, K.S. (eds). Springer- Verlag. Berlin, pp: 150-187.
16. **Mahmoud A M Gadel-Rab A G and Shawki N A (2017):** Effect of different feeding behaviors on the roof of buccal cavity of two bird species. Egypt. J. Zool., 67:175-190.
17. **Fahrenbach W H and Knutson D D (1975):** Surface adaptation of the vertebrate epidermis to friction. J. Invest. Dermatol., 65**:** 39-44.
18. **Shawki N A and Ismail A I (2006):** Functional morphology of the lingual apparatus of the common moorhen, *Gallinula choropus* Meridionalis (Aves: Rallidae). Egypt. J. Zool., 46**:** 41-57.
19. **Sperry D G and Wassersug R J (1976):** Proposed function for microridges on epithelial cells. Anat. Rec., 185**:** 253-258.
20. **Iwasaki S and Kobayashi K (1986):** Scanning and transmission electron microscopical studies on the lingual dorsal epithelium of chickens. Acta. Anat. Nippon., 61**:** 83-96.
21. **Jackowiak H and Godynicki S (2005):** Light and scanning electron microscopic study of the tongue in the white-tailed eagle (*Haliaeetus albicilla*, Accitripidae, Aves). Ann. Anat., 187**:** 197-322.
22. **Bhattacharyya B N (1990):** The functional morphology of the lingual apparatus of two species of imperial pigeons, *Ducula aenea Nicobarica* and *Ducula badia Insignis*. Proc. Zool. Soc. Calcutta, 43**:** 65-93.
23. **Garqiulo A M Lorvik S Ceccarelli P and Pedini V (1991):** Histological and histochemical studies on the chicken lingual glands. Brit. Poult. Sci., 32**:** 693-702.
24. **Homberger D G (1982):** Hydraulic structures in the avian lingual apparatus. Am. Zool., 22**:** 943-954.
25. **Alexander R M (1968):** Animal Mechanics. University of Washington Press. Seattle. Publ. U S A., Chap., 5**:** Pp: 152-162.