Morphological characterization of hard palate in the Tabby cats

Caracterizacion morfológica del paladar duro en los gatos Tabby

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ABSTRACT

The aim of this study was to carry out the morphological characterization of the Tabby cat's hard palate by light and scanning electron microscopy (SEM), in addition to gross and morphometric analysis. A total of 20 Tabby cat heads used. The materials was provided from the Balıkesir Metropolitan Municipality Street Animals Temporary Nursing Home and Rehabilitation Center, Balıkesir, Turkey. Adult, regardless of gender difference, died from variety of reasons and 20 Tabby cat cadavers which weigh approximately 3-4 kg were brought to the laboratory in accordance with the procedure. Hard palates were subjected to gross examination, morphometric analysis, light and SEM. The Tabby cats hard palate was split up rostral and caudal parts and incisive papilla (papilla incisive), rugae palatine and with a gutter between them, the presence of palatine raphe and secondary rugae palatine were observed. The rostral and caudal parts of the hard palate were determined to have a smooth appearance with SEM 540X magnification and the presence of epithelial desguamations was found out. It was determined that the rostral and caudal part of the Tabby cat's hard palate had a honeycomb appearance of the microplicae of the 3000X magnification hard palate epithelium with SEM. Round islets were detected with the SEM at the magnification of 600X of the incisive papilla, the 270X of the rostral-caudal parts and the 44X of the gutter between the palatine rugae. These adaptations of the Tabby cat's hard palate might increase its efficiency during ingestion and mastication of hard food and in directing the food backward. The study shows the detailed anatomical description of the hard palate of the Tabby cat with light and SEM.

Key word: Hard palate; SEM; Tabby cat

RESUMEN

El objetivo de este estudio fue investigar la morfología del paladar duro del gato Tabby mediante microscopía óptica y electrónica de barrido (SEM), además de análisis macroscópico y morfométrico. Se utilizaron un total de 20 cabezas de gato Tabby. Los materiales fueron proporcionados por el Centro de Rehabilitación y Hogar de Ancianos Temporales de Animales Callejeros del municipio Metropolitano de Balıkesir, Balıkesir, Turquía. Los adultos, independientemente de la diferencia de género, murieron por diversas razones y se llevaron al laboratorio 20 cadáveres de gatos atigrados que pesaban aproximadamente 3-4 kg de acuerdo con el procedimiento. Los paladares duros se sometieron a examen macroscópico, análisis morfométrico, microscopía óptica y SEM. El paladar duro de los gatos Tabby estaba dividido en partes rostral y caudal y papila incisiva palatine rugosa y con una canaleta entre ellos, la presencia de rafia palatine y secundaria rugae palatine fueron observados. Se determinó que las partes rostral y caudal del paladar duro tenían una apariencia lisa con SEM 540X de aumento y se encontró la presencia de descamaciones epiteliales. Se determinó que la parte rostral y caudal del paladar duro del gato Tabby tenía una apariencia de panal de abeja de las microplicas del epitelio del paladar duro de aumento 3000X con SEM. Los islotes redondos se detectaron con el SEM al aumento de 600X de la papila incisiva, el 270X de las partes rostral-caudales y el 44X de la canaleta entre las rugosidades palatinas. Estas adaptaciones del paladar duro del gato Tabby podrían aumentar su eficiencia durante la ingestión y masticación de alimentos duros y al dirigir los alimentos hacia atrás. El estudio muestra la descripción anatómica detallada del paladar duro del gato Tabby con microscopía óptica y SEM.

Palabras clave: Paladar duro; SEM; gato atigrado



INTRODUCTION

The Feline family consists of 39 felines and these cat breeds descend from the last common ancestor, '*Pseudaelurus*' who lived 10–15 million years ago. The Anatolian cat, which is a member of '*Felidae*' is an extension of '*Felislybica*' which evolved 3–4 million years ago from its last common ancestor, the wild cat [1]. It has been observed that the '*Felis silvestrislybica*' whose archaeological remains were analyzed by ancient deoxyribonucleic acid (DNA), contributed to the gene pool of different historical periods in the Near East and Egypt populations [2].

Tabby cats (*Felis catus*) constitute the majority of the cats kept at home in Turkey and the name Tabby is quite common. Tabby cats, also called stray cats, make up the majority of street cat species in the Country. The Tabby cat is used as a noun in English- speaking Countries, Tabby, which expresses a skin pattern, is based on the word tigris, which has its etymological origin in ancient Greek and Latin languages meaning tiger. Today, Tabby is used sense reminiscent of a tiger sriped back (*Panthera tigris*). The story of the Tabby cat, which is expressed as 'Chattigre' in French and 'Gattotigrato' in Italian and 'Tigerkatze' in German, is slightly different. While these cats were called tigercats in England, the name of fabric impotent from the Attabiye District of Baghdad, which is known for weaving taffeta silk fabric in a way that resembles the lines of a tiger, became heavy and as a result of this story, it began to be called Tabby [<u>3</u>].

Taste buds are responsible for perceiving the sense of taste. In mammals, most of the taste buds in the tongue, soft palate, epiglottis epithelium, larynx, and the retromolar mucosa are located on the tongue papillae [4, 5].

The sense of taste, which functions together with smell, functions both peripherally and centrally in physiological harmony. The sense of taste, which functions together with smell, is a physiological harmony peripherally at the reseptor level and centrally at the reticular activating level [6]. This union leads us to the concept of taste. The sense of taste in mammals with four primary sensations; salt and its stimulant NaCl; sour and its stimulant kşi acetic acid; sweet and its stimulant sucrose; spicy and its stimulant quinine. In mammals, taste buds embedded in the epithelium of the tongue are small structures (20–42 μ m) and contain between 50 and 150 taste receptors.

In this research, macro-anatomical evaluation and morphometric measurements of the hard palate of the Tabby cat, which is common in Turkey, were made; Tissue samples taken from the anatomical formations on the hard palate were examined with SEM (Jeol, model JCM-5000 NeoScope™ Jeol Ltd., Tokyo, Japan), and light microscopy. (Nikon, Eclipse, E 200 MB-R, Tokyo, Japan)With data obtained, it is aimed to determine the morphological and morphometric characteristics of the hard palate of the Tabby cat and to determine the similarities and differences with other domestic mammal species and humans in the light of current literature. In addition, knowing the hard palate anatomy of Tabby cats is important in clinical examination in the detection of structural disorders such as congenital-acquired anomalies.

MATERIALS AND METHODS

The materials used in the thesis study were obtained from Balıkesir Municipality Stray Animals Temporary Care Home and Rehabilitation Center with permission. The research material consisted of 20 Tabby cat cadavers, which were adults, died for various reasons, without gender discrimination, and weighed approximately 3–4 kg. The study was carried out with the approval of Balikesir University Animal Experiments Local Ethics Committee (Decision no: 2020/2–8).

Cat cadavers were transferred from Balıkesir Metropolitan Municipality Stray Animals Temporary Care and Rehabilitation Center to Balıkesir University Veterinary Faculty Anatomy Application Laboratory. To expose the hard palate of the cadavers, the maxillary and mandibular jaws were separated from each other by making an articulatio temporomandibularis incision. The hard palate upper jaw parts are numbered and fixed with 10% formaldehyde. After fixation, the hard palate was washed and photographed with a Canon 50 Digital camera (Ota, Tokyo, Japan). The macro anatomical nomenclature of hard palate structures was made according to Nomina Anatomica Veterinaria (2017)[7].

Morphometric measurements of different parts of the hard palate were made with a digital caliper (Piranha PDC 1850 Digital Caliper, China). The total length of the hard palate, the length of the narrow (rostral) and wide (caudal) segment, the length of the rough and straight segment, the width of the narrow and wide segment, the total length of the palatine raphe, the length of palatine raphe deep and shallow segment, number of palatines rugae (double), and the incisive papillae diameter was measured. Data were expressed by calculating the mean and standard deviation values in millimeters (mm).

Scanning electron microscopy (SEM)

Samples taken from the hard palate were primarily fixated in glutaraldehyde solution at +4°C for 24 h. The sample was kept in 0.2 M phosphate buffer (pH 7.2) for 15–30 min, for a total of 1 h at +4°C, by changing the solution 2 times. The samples removed from the phosphate buffer were kept in the solution prepared by putting 1% osmium tetraoxide in 0.2 M phosphate buffer (pH 7.2) and kept for 1 h at +4°C for secondary fixation. Afterward, the samples were kept in a phosphate buffer of 0.2 M (pH 7.2) for a total of 1 h. After the samples were subjected to dehydration at 30–60–70–100% levels for 15 min, they were kept in hexamethyldisilazane and dried using the critical dried point method [$\underline{8}$]. Samples; after drying, it was covered with gold and examined with a NeoScope JCM–5000 scanning electron microscope (Tokyo, Japan).

Light microscopy

Samples obtained from the hard palate fixed with 10% formaldehyde were placed in tissue cassettes in 0.3 cm sections. Blocking was performed by passing the tissue cassettes through formol-alcoholxylol- paraffin solutions. After paraffin sections were cut (Leica Biosystems RM2245, GmbH, Wetzlar, Germany) at 5 µm thickness and placed on the slide, they were kept in the oven at 60-70 °C until the paraffin melted. It slowly passed through the previously prepared xylol solution. Sections were passed through alcohol solutions of different percentages. The sections were completely freed from paraffin by washing them with distilled water. Subsequently, these tissues were stained with Hematoxylin-Eosin. After staining, the tissues were washed with tap water. It was then passed through a 70-99.9% alcohol solution. The prepared slides were kept in xylol for at least 10 min. The coverslips were covered and examined under the microscope. The prepared slides were examined under Nikon, Eclipse light microscope (E 200 MB-R, Tokyo, Japan).

RESULTS AND DISCUSSION

Gross morphology

The total length of the hard palate of the cat, consisting of rostral and caudal parts was 28.11±2.4 mm. The total length of the hard palate consisted of 72.92% caudal parts and 27.08% rostral parts (TABLE I).

The width of the rostral part at the level of the premolar tooth was 16.25 ± 2.25 mm, and the width of the caudal part between the second premolar tooth and the last molar tooth was 25.95 ± 2.4 mm. The deep part of the palatine raphe was between the incisive papilla and the last molar tooth and its length was 23.40 ± 1.8 mm. The length of the shallow section behind the last palatine rugae was 2.54 ± 0.6 mm, 6-8 palatine rugae with transverse extension was seen on both parts of the palatine raphe.

<i>TABLE I</i> Hard palate morphometric analysis		
Hard Palate Parameteres	x ± SD (mm)	
Total length of hard palate	28.1±2.4	
The length of narrow (rostral) part	7.6±1.8	
The length of wide (caudal) part	20.5±2.3	
The length of rough part	20.6±2.9	
The length of flat part	7.4±2.6	
The widht of narrow (rostral) part	16.2±2.2	
The widht of wide (caudal) part	25.9±2.4	
The total length palatina raphe	25.9±1.9	
The length of the deep part of the <i>raphe palati</i>	23.4±1.8	
The length of the shallow part of the <i>raphe palati</i>	2.5 ± 0.6	
Diameter of incisive papilla	2.4±0.4	

x: Average, SD: standard deviation

Palatine rugae was seen in the entire rostral part of the hard palate, but only in the anterior of the caudal part. The hard palate was divided into rough and flat parts according to the palatine rugae distribution. The rough part between the incisive papilla and last palatine rugae constituted 73.46% of the length of the hard palate. The flat part between the end of palatines rugae and the level of the last molar tooth constituted 26.54% of the hard palate. The total length of the feline hard palate consisted of 37.7% was a narrow piece and 62.3% was a wide piece.

The incisive papilla was located between the first palatine rugae and incisive teeth. The diameter of the incisive papilla, which was round was round like a rasory and had an olive-like shape (FIG. 1), was measured at 2.4 ± 0.4 mm.

The hard palate had transversely elongated palatine rugae characterized by mucosal folds and a median shallow groove connecting them to the midline. It was determined that the median groove, called palatine raphe, extends from the incisive papilla to the level of the last molar tooth and had two parts (FIG. 1). The deep part between the anterior part of the last molar and the incisive papilla was determined as the shallow part behind the last molar tooth.

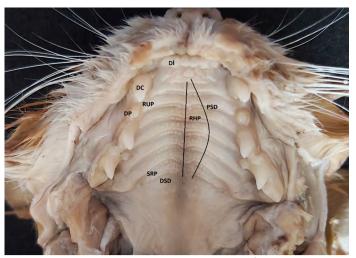


FIGURE 1. Photograph of the hard palate of the Tabby Cat: Dİ: D. Incisivi, DC: Canin, DP: D. Premolar, Pİ: Incisive Papilla, RHP: Palatine Raphe, RUP: Palatine Rugae, PSD: Rough Hard Palate, DSD: Straight Hard Palate, SRP: Seconder Palatine Rugae

Palatine rugae on both sides of the palatine raphe had a symmetrical appearance. 7 palatine rugae in 19 cats, 8 palatine rugae in 1 cat and secondary papilla (palatine rugae) in 17 cats were determined (TABLE II).

The seconder palatine rugae located between the last palatine rugae and the last molar tooth and had an asymmetrical appearance. The mean number of palatine rugae was 7.05 ± 0.22 and the seconder papilla was not included in this number. The mean of the length measurement of the palatines rugae was calculated as 3.49 ± 0.42 mm on the left and 3.54 ± 0.42 mm on the right. Palatines rugae from rostral to caudal lengths increased (TABLE II).

The mean width measurement of the palatines rugae was 1.84 ± 0.22 on the left and 1.84 ± 0.23 mm on the right. Although the width measurements on both sides of the palatine raphe show slight differences, they showed a symmetrical feature. The distance of the furrows between each palatine rugae was determined symmetrically. Palatine raphe, which was not prominent until the fifth rugae palatine, was prominently detected from the end of the fifth palatine rugae.

<i>TABLE II</i> Palatinae rugae morphometric analysis		
of palatine rugae	x ± SD (mm)	
Left	7.05±0.22	
Right	7.05±0.22	
Left	3.494±0.42	
Right	3.542 ± 0.42	
Left	1.844±0.22	
Right	1.845±0.23	
	e morphometric ana of palatine rugae Left Right Left Right Left Left	

x: Average, SD: standard deviation

Scanning electron microscopic examination

C-shaped incisive papilla starting from dentes incisive to caudalaterally, smooth at low magnification (FIG. 2a), epithelial desquamations at high magnification (FIG. 2b), microplicae with honeycomb appearance (FIG. 2c), the fiber in epithelial layers, connective tissue bundles and round islets were seen (FIGS. 2d and 3a). Heterogeneously distributed, round islets of different sizes were determined on the incisive papilla with a keratinized structure (FIG. 3b).

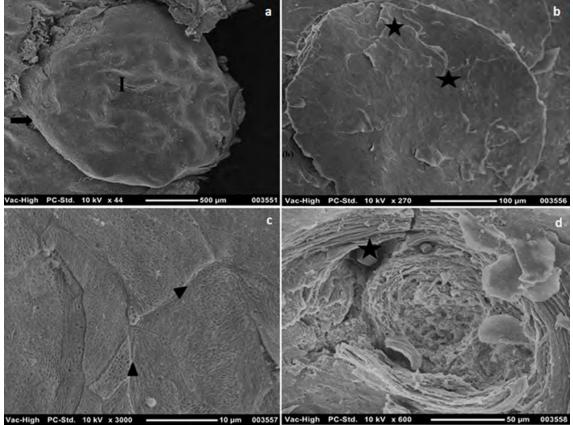


FIGURE 2. The appearance of the incisive papilla in the scanning electron microscope. Incisive papilla (I); a: 44X magnification, demarcated groove on both caudal-lateral sides (arrow), b: appearance of keratinized epithelial desquamations (star) 270X magnification, c: epithelium of the papilla incisiva divided by secondary septa (arrow head) 3000X magnification, d: epithelial and submucosal layers of the incisive papilla, round islet (star) 600X magnification

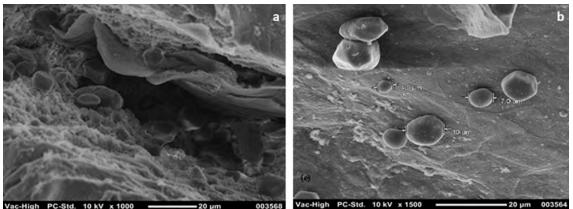


FIGURE 3. View of the rostral and caudal part of the Tabby hard palate in scanning electron microscopy. a: round islets and connective tissue 1000X magnification, b: islets diameters 4.5 μm, 7 μm, 10 μm. 1500X magnification

It was observed that the transversely arranged palatines rugae were conical in shape, free at the tip, and connected to each other at the base (FIG. 4a).

In the image taken from the groove between the palatine raphe and the palatine rugae, heterogeneously distributed round islets of different sizes were seen (FIG. 4b).

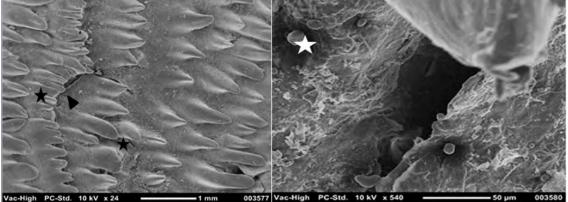


FIGURE 4. The image of the palatine raphe and the groove between the two palatines rugae of the hard palate of the Tabby cat in scanning electron microscopy. a: rugae palatina (star), groove between two rugae palatine ridges (arrow head) 24X magnification, b: desquamation of epithelial layers and round islet (star), connective tissue bundles and microplicae of epithelial layers 540X magnification

Light microscopic examination

Complementary epithelium, c-shaped hyaline cartilage, artery, and nerve plexus were seen in the incisive papilla image of the hard

palate (FIG. 5a). Epithelium in the caudal part of the hard palate and in the palatine raphe; It was a keratinized structure, and was a deeper stratified squamous epithelium. Microscopic papillae, arteries and veins were seen in the lower layer of the epithelial layer (FIG. 5b).

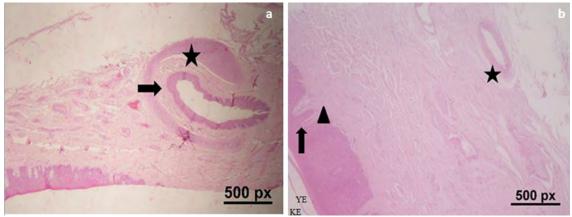


FIGURE 5. Image of incisive papilla of Tabby hard palate in light microscope. a: hyaline cartilage (star), complementary epithelium (arrow) 10X magnification, b: Light microscope image of cat palatine raphe; Keratinized epithelium (KE), stratified squamous epithelium (YE), microscopic papillae (arrow head), artery (star), vein (arrow) 10X magnification

In agreement with the presented study; It has been reported that the hard palate was divided into rostral and caudal parts in Anatolian lynx (*Lynx lynx*) [9], Little Indian Civet (*Viverricula malaccensis baliensis*)[10], rabbit (*Oryctolagus cuniculus*)[11] and Egyptian goat (*Nubian ibex*)[12]. The rostral portion, which was the section between the incisive papilla and premolars; The caudal part, which started to expand from the premolar tooth level to the caudal; ends at the level of the molar teeth [9, 10, 11, 12].

On the other hand, the width of the hard palate in Sambar deer (*Rusa unicolor*)[<u>13</u>] and cattle (*Bos taurus*)[<u>14</u>] was wide in the rostral and caudally and narrow in the middle. Short-tailed monkey hard palate; rostral and caudal widths were equal to each other [<u>15</u>]. In the squirrel (Sciuridae) hard palate; width measurements at the end, middle, and terminal were close to each other [<u>16</u>]. These statements were not similar to the presented study.

Hard palate length was measured at 26.4 ± 1.27 cm in cattle [14], 15.80 \pm 0.47 cm in porcupines (*Erethizon dorsatum*) [17], 13 cm in Sambar deer [13] and 11.80 \pm 1.05 cm in Bakerwali goat [18]. Measurement of the length of the hard palate of the cat present study was measured as 28.1 ± 0.24 mm, unlike in these studies.

Palatine raphe on the hard palate had a caudally oriented and serrated appearance, symmetrically and transversely aligned, on both sides of the palatine rugae in the study. Palatine raphe was not prominent in all cats until the 4th rugae palatine, but had a more shallow appearance, deepening from the end of the 4th palatine rugae. The mean number of palatine rugae on the hard palate, being equal on the right and left side was 7. Except for an average of 7 palatine rugae, 1 secondary palatine rugae was seen in 17 cats. The secondary palatine rugae were located between the last palatine rugae on the hard palate and the last molar tooth level. The secondary palatines rugae on the hard palate of two Tabby cats with pigmented features did not show symmetrical features. The number of palatine rugae was determined at 8–22 in rabbits [11] and in the fur animal rabbit, which is a mixture of rabbits and squirrels; 4–6 [19], 9–10 in the Small Indian Civet [10], 9 in the African giant mouse (Rhabdomys) [20], 6-10 in the dog(Canis familiaris)[21], 6-9 in the Anatolian lynx[9], 7 in the squirrel [16] and 8 pieces in the short-tailed monkey (Pigtail Macague) [15]. The number of palatine rugae was similar to those in the Anatolian lynx [9], dog [21], and squirrel [16], in the present study.

In presented study, palatine raphe, which became evident after the end of the 5th palatine rugae; In the studies of the Little Indian Civet [10] and the short-tailed monkey [15], it was similar to the fact that the palatine raphe was not prominent until the 5th palatine rugae.

The transversely aligned palatines rugae differ in the Little Indian Civet [10], they were prominent in the front and, not in the back, but were prominent in the front and back in the presented study.

Similar to presented study; in Anatolian lynx [9] and squirrels [16] had the longest palatine rugae on both sides of the palatine raphe, where the hard palate meet the smooth part, and at caudally there were a secondary palatine rugae.

In a study by Toda [22], the palatine rugae on the hard palate of the cat were transversely aligned, and caudally curved, and the anterior slope of each back was longer than the posterior slope, and the posterior slope was slightly steeper, similar to the present study [22]. Against this; a study by Yamazaki *et al.* reported that the crocodile (*Crocodylus*) hard palate did not have palatine rugae as in cats, mice (*Mus musculus*), humans and other animals and that the crocodile had many papilla-like conical projections on the hard palate were not compatible with the presented study [23].

In this study, based on the examination of the hard palate of the Tabby cat, the incisive papilla was in the shape of a rosary and had an olive-like appearance due to its narrowing on the sides. Similarly to the study; the hard palate incisive papilla of goat [24], dog [20] and Indian Civet [10] had a round shape. Against this; incisive papilla was found on the hard palate in a triangular pyramid shape of the African giant mouse [20], half-moon shape in Bakerwali goat [18], diamond-shaped in Sambar deer [13], and as a small carboy in Anatolian lynx [9].

The similarity with the presence of round islets in SEM images of the human hard palate, differs from the light microscope image. In the research of Imfeld and Schroeder [25], where the islets were located was the middle, posterior part of the soft palate and border with the hard palate. The present study was similar to the presence of round

islets in the grooves of the palatines rugae, incisive papilla, palatine raphe, and palatine rugae grooves like in humans [25] and Anatolian lynx [9] hard palate studies with SEM. It differed from these studies carried out under the light microscope because the presence of islets in the hard palate with a keratinized structure was not mentioned.

The results of the stratum corneum with a keratinized structure and the fact that it is covered with stratified squamous epithelium, obtained in histological examination of the hard palate with light microscopy, were similar to Anatolian lynx [9], water buffalo [26], Egyptian goat [12], goat [24], crocodile [23] and human [27, 28] studies.

In the histological examination of the hard palate of rabbit [29] and cat [22], the presence of vascular arterial and venous plexus was similar to the findings of the presented study. In addition, the incisive papilla containing 'C' shaped hyaline cartilage and the presence of nerves and vessels was similar to those that Anatolian lynx [9] reported.

CONCLUSION

It was concluded that the shape and direction of the rugae palatines and the raphe palatine ensure the backward orientation of food. Taste bud-like round islets were observed as taste-related anatomical structures in the samples examined by Scanning electron microscope. The findings obtained in the study were discussed in terms of similarities and differences with the findings in studies on humans, domestic and wild animals. It was thought that the findings obtained in the study will contribute to the literature and will shed light on the studies to be done in this field.

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The preprint of this study is available at https://www.authorea.com. (https://www.authorea.com/doi/full/10.22541/au.167355886.63514200).

Conflicts of interest

The authors declare no potential conflict of interest.

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