

## The Light and Scanning Electron Microscopic Structure of the Papilla Vallatae in the Porcupine (*Hystrix cristata*)

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**Abstract:** This study was aimed to examine detailed structure of papilla vallata in porcupines by light and scanning electron microscopy. For this purpose, 5 mature porcupines (3 males and 2 females) were used. Papilla vallata was one each in both side of median line in dorsal surface of tongue base. There was taste buds (Calculus gustatorius) only inner wall of ditch of papilla. Maximum 9 taste buds were counted on one side. The mean surface area of papilla ditch wall was calculated as 30.26 mm<sup>2</sup>. In addition, mean micrometric measurements of papilla vallata were width of papilla (3225 µm), length of papilla (1875 µm), depth of ditch (1890 µm), length of the taste bud (206.25 µm), width of the taste bud (117.86 µm) and epithelial thickness (362.5 µm). Two types of taste bud cells that were lightly dyed (Epitheliocytus sensorius gustatorius) and densely dyed (Epitheliocytus sustentans) were observed as arranged vertically through depth of ditches. The borders of surface epithelial cells of papilla vallata were clearly observed on scanning electron microscope. There were micropits and microridges on cell surfaces. In conclusion, papilla vallata of porcupines were examined in detail by light and scanning electron microscopy and then the similarities and differences with other mammals were determined.

**Key words:** Porcupine, papilla vallata, scanning electron microscopy, taste buds, mammals, Turkey

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### INTRODUCTION

The rodents (Rodentia) which are known the widest order of placental mammals, comprise the largest and the most diverse group of mammals with over 1700 species present day. Porcupine is from Hystricidae family which constitutes a small group of the order Rodentia (Weichert, 1970; Besselsen, 2002; Atalar *et al.*, 2004; Atalar and Yilmaz, 2005; Tarakci *et al.*, 2007; Yaman *et al.*, 2007).

Although, some morphologic studies on the organs and tissues of the porcupine (Atalar *et al.*, 2003; Atalar and Yilmaz, 2004a, b; Ozdemir *et al.*, 2005, 2006) have been investigated and information about studies on the papilla vallata has been reported inadequate in the literature. In addition to, macro and micro-anatomical studies were present some wild species (Atalar and Yilmaz, 2004a, b; Timurkaan *et al.*, 2006; Atalar *et al.*, 2007, 2009; Karan *et al.*, 2007, 2008).

There are swellings called *papilla lingualis* on the tongue that is an organ in the muscle structure. These papillae are two types called as mechanic and gustatoric. Papilla vallata which is a *gustatoric papilla* has located to radix linguae. The macro and micro-anatomical structures

of these papilla show differences according to the animal species (Banks, 1986). The rodents (Rodentia) which are the widest order of placental mammals, comprise more than half of the mammals know present day. The porcupine belongs to the Hystricidae family which constitutes a small group of the order rodentia (Atalar *et al.*, 2003; Atalar and Ceribasi, 2006; Cigremis *et al.*, 2008; Atalar *et al.*, 2009).

There was relatively more information on papilla vallata of various vertebrates including several species and strains of rodents (Cano *et al.*, 1978a, b; Crossmon, 1937; Emura *et al.*, 1999; Gude *et al.*, 1982; Hosley and Oakley, 1987; Iwasaki *et al.*, 1988, 1996, 1997; Kobayashi, 1990; Miller and Smith, 1984). However, the literature investigation showed that there was no information on light and scanning electron microscopic structure of papilla vallata in porcupine. There is only one neurohistological study on the tongue of the porcupine (Kubota *et al.*, 1966).

The objective of the present study has been to investigate the light and scanning electron microscopic structure of papilla vallata in porcupines for the first time to extend the knowledge in this field.

**MATERIALS AND METHODS**

In this study, five adult (3 males and 2 females) porcupines of different ages and hunted by villagers in Eastern Anatolia (Turkey) were used. Deep anaesthesia of animals was induced by initial injection of ketamine HCl (Ketone 10-15 mg kg<sup>-1</sup> i.m.) followed by xylazine HCl (Rompun 0.10-0.15 mg kg<sup>-1</sup> i.m.) (Atalar *et al.*, 2003; Atalar and Yilmaz, 2004b; Yaman *et al.*, 2008). For the light microscopic examination, area of vallat was fixed in 10% formalin solution. Papillae samples were processed routinely for paraffin embedding and sectioned at 5-7 µm. Sagittal sections from papillae were stained by hematoxylin eosin (Luna, 1968). Micrometric measurements were performed by an ocular micrometer.

The tongue samples of five porcupines were used for the scanning electrone microscopic examination. These tongue samples were used as a whole and all samples were rinsed with the physiologic water-buffered saline. Tissue samples were prefixed in 3% gluteraldehyde (pH: 7.3) solution with phosphate buffer. After tongue fixation in the same buffer, samples were post-fixed in 1% osmium tetra-oxide at 37°C for 1.5 h.

Then, the tongue samples were treated by 3N hydrochloric acid at 60°C for 20 min to remove the mucus on the surface of the tissue. The tongue samples were then rinsed with alcohol and amyl-acetate solution and dred by using critical-point-dryer. Finally, they were coated with the gold by using VG Microtech Polaron SC 500 sputter-coater under 0.05 Torr pressure. All samples were examined, photographed and evaluated by the Jeol JSM 5600 scanning electron microscope between 5 and 15 kV.

**RESULTS AND DISCUSSION**

It was found that the mean length and wide of porcupine tongue were 6.7 and 1.6 cm, respectively. Sulcus medianus was deep in the front 1/3 ratio, less clear in the middle 1/3 ratio and unclear in back 1/3 ratio of tongue (Fig. 1).

It was notified that the number of papilla vallata was three on the porcupine tongue. Papilla vallata was two with only one in the both sides of median line in the dorsal surface of tongue. Both sides and back of it was surrounded with ditches.

Micrometric values belong to papilla were shown in Table 1. Taste buds were found 206 µm in length and 117 µm in width in the porcupine and the area of the wall

of papilla ditch was calculated as 30.26 cm<sup>2</sup> in average. Papilla was covered with a multi layered and flat epithelium. Epithelial thickness was measured from the region with taste buds. Epithelium on the dorsal of papilla was very variable. Taste buds were only present in the inner wall of papilla ditch. Maximum 9 taste buds could be counted on one side. These taste buds had a clear taste porus (*Porus gustatorius*). Von Ebner gland was present and opened to the base of ditches with a channel (Fig. 2-4).

It was clearly observed on scanning electron microscope examination that all around the papilla vallata was surrounded by the ditch on the both side and back of it (Fig. 5). Two type taste bud cells that were lightly dyed and densely dyed were determined as arranged vertically



Fig. 1: View of the tongue of porcupine. pv = papilla vallata

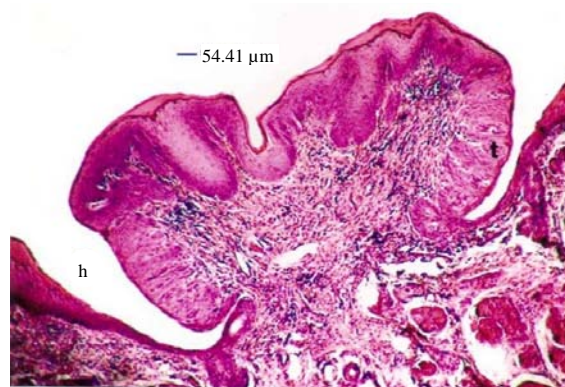


Fig. 2: General view of papilla vallata from vertical cross-section. t = taste bud, h = Ditch

Table 1: Avarage micrometric measurements of papilla vallata

Papilla vallata	Width of papilla	Lenght of papilla	Depth of ditch	Lenght of taste bud	Width of taste bud	Epithelial thickness
Avarage measurement (µm)	3225±70	1875±183	1890.0±21.6	206.25±25.00	117.86±12.00	362.5±68.0

Values are given as X±SD

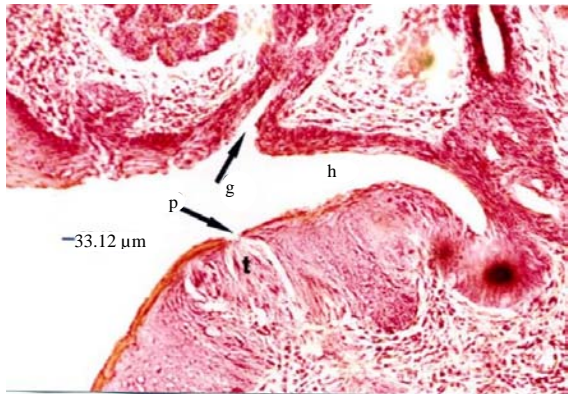


Fig. 3: View of the base of ditch. t = taste bud, h = Ditch, p = porus of ditch, g = canal of glandula gustatori

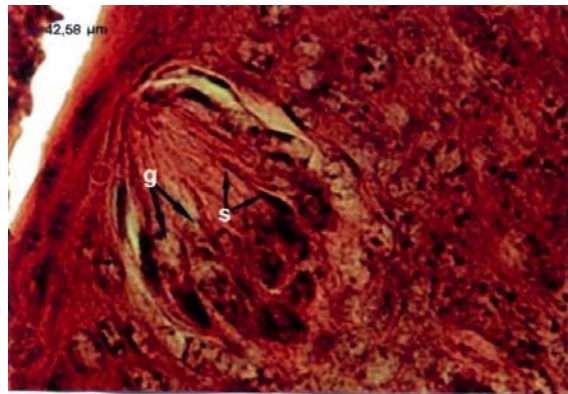


Fig. 6: View of taste bud cells. g = Light (Gustatorius) cells, s = Dens (Sustentans) cells



Fig. 4: General view of papilla vallata from horizontal cross-section. t = taste bud, h = ditch

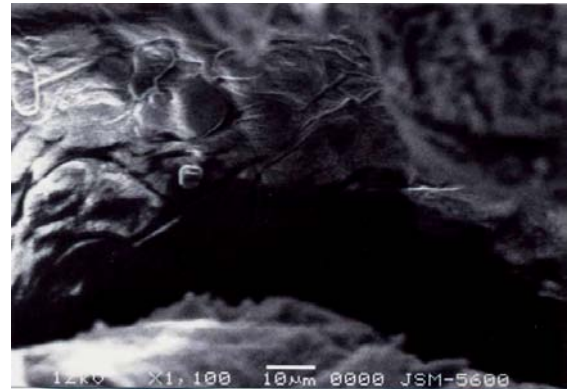


Fig. 7: Entrance of papilla vallata ditch



Fig. 5: Superficial view of papilla vallata

through depth of ditches (Fig. 6). It was found that these cells were not completely filled to the taste buds and

occupied nearly 2/3 of them. The borders of surface epithelial cells were determined by doing higher zoom on the papilla surface. There were micropits and microridges on the surface of these cells (Fig. 7 and 8).

The findings such as possessing a connective tissue and covered a very flat epithelium of papilla vallata notified by others (Agungpriyono *et al.*, 1995; Krause and Cutts, 1982) were also observed in the present study similarly. Surface epithelial of papilla were examined by doing higher zoom and showed cells very clearly (Iwasaki *et al.*, 1988, 1996, 1997). There were micropits and microridges on the surface of these cells as well (Iwasaki *et al.*, 1997).

It was notified that the number of papilla vallata was one in mouse (Kinnamon *et al.*, 1985; Utiyama *et al.*, 1995) in hamsters (Miller and Chaudry, 1976; Miller and Smith, 1984) in rats (Toyoshima and Shimamura, 1979), two in porcupine (Kubota *et al.*, 1966), three in Japanese dormouse (Kubota and Togawa, 1966) and in flying

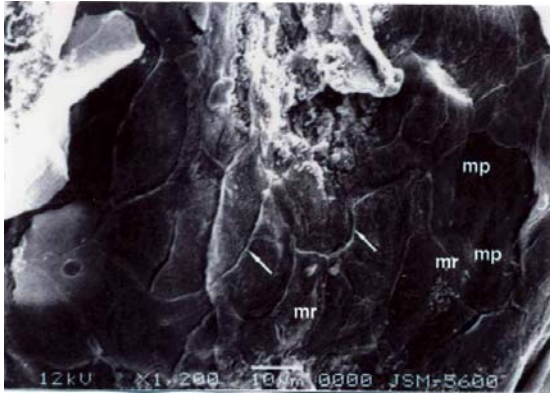


Fig. 8: High magnified view from surface of papilla vallata. mp = micropit, mr = microridge, arrows = Cell borders

squirrel (Emura *et al.*, 1999). The finding of the present study showed similarity by that of Kubota *et al.* (1966) on the porcupine.

Taste buds were 106  $\mu\text{m}$  in length and 59  $\mu\text{m}$  in width (Hosley and Oakley, 1987) or 81  $\mu\text{m}$  in length and 45  $\mu\text{m}$  in width in rats, 46  $\mu\text{m}$  in size (Mistretta and Baum, 1984) in hamster. In the present study, taste buds were found 206  $\mu\text{m}$  in length and 117  $\mu\text{m}$  in width in the porcupine.

The surface area of ditch wall was given as 2.42  $\text{mm}^2$  or 3.67  $\text{mm}^2$  (Hosley and Oakley, 1987) in rats and 0.83  $\text{mm}^2$  in mouse. It was found as 30.26  $\text{mm}^2$  in the porcupine.

It has been reported that taste buds of papilla vallata have possessed 4 type cells (Cano *et al.*, 1978a, b), 3 type cells (Banks, 1986) and 2 type cells (Gude *et al.*, 1982). In the present study, 2 type cells were observed.

It was reported that taste buds intra-epithelial arranged both side walls of ditch in mouse (Kinnamon *et al.*, 1985), rats (Oakley, 1974; Toyoshima and Shimamura, 1979) and hamsters (Miller and Chaudry, 1976; Miller and Smith, 1984). Otherwise, in the present study taste buds were only observed in the inner wall of ditch in the porcupine.

## CONCLUSION

The structure of papilla vallata of porcupine was detail examined for the first time by light and scanning electron microscopy. Findings of the present study will contribute important information for this study area.

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