Morphology and structure of filiform papillae of neonates: a light and scanning electron microscopic study

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شكل وتركيب الحلمات الخيطية في الولدان: دراسة بالمجهر الضوئي والمجهر الإلكتروني التفرسي راجية البرنس حسين نور الدين

خلاصة : تهدف هذه الدراسة إلى وصف شكل وتركيب الحلمات الخيطية في الولدان. أخذت مشر عينات هيستولوجية من الجزء الأمامي للسان في عشرة من ولدان ولدوا بعد موتهم داخل الرحم. وتم تثبيت العينات وتجهيزها مخبرياً للفحص بالمجهر الضوئي والمجهر الإلكتروني التفرسي. وقد بدت الحلمات الخيطية في شكل نتوءات صغيرة إصبعية الشكل ذات أطراف مستديرة، كما ظهر أن كل حلمتين أو ثلاث حلمات تنشأ من قاعدة واحدة قبل أن تأخذ في الانفصال عن بعضها البعض. ووجد أن الظهارة المغطية للحلمات كانت شبه متقرنة على علاف الظهارة بين الحلمات.

ABSTRACT The present study was carried out to describe the morphology and structure of filiform papillae of the neonate. Ten tissue specimens taken from the anterior portion of the tongue of ten stillborn neonates were fixed and processed for both light and scanning electron microscopy. Filiform papillae appeared as small finger-like projections with rounded tips and appeared to develop as two or three papillae originating from one base; subsequent splitting of the papillae from each other then took place. The epithelium covering the papillae was parakeratinized while that covering the interpapillary regions was nonkeratinized.

Morphologie et structure des papilles filiformes des nouveau-nés: étude au microscope optique et au microscope électronique à balayage

RESUME La présente étude a été réalisée pour décrire la morphologie et la structure des papilles filiformes des nouveau-nés. Dix échantillons de tissus prélevés sur la partie antérieure de la langue de dix enfants mort-nés ont été fixés sur des lames et examinés au microscope optique et au microscope électronique à balayage. Les papilles filiformes sont apparues comme de petites saillies digitiformes aux bouts arrondis qui se ramifient en deux ou trois papilles à partir de la même base d'origine pour se séparer ensuite. L'épithélium qui couvre les papilles était parakératinisé tandis que celui qui couvre les régions interpapillaires n'était pas kératinisé.

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Introduction

The mucosa of the dorsal surface of the tongue is unlike anywhere else in the oral cavity. Although covered by what is functionally called a masticatory mucosa, it is a highly extensible lining and has different types of lingual papillae [1].

Filiform papillae cover the entire anterior part of the dorsum of the tongue and consist of cone-shaped structures, each with a core of connective tissue covered by keratinized epithelium. Together they form a tough abrasive surface which is involved in mastication. With scanning electron microscopy, filiform papillae of the human tongue have been seen to contain 5–12 hairs which are covered with a massive plaque of microorganisms [2]. The surface of the body of the papilla is smooth and covered by epithelial cells that has a honeycomb pattern with prominent overlapping cell boundaries [2].

Many previous studies have been conducted to describe the age-related changes in the human tongue. These have revealed atrophy and disorganization of the tongue papillae and gradual loss of taste buds [3], with increased collagen fibre content and fat infiltration in the lamina propria [4]. Very few studies have been carried out on the development of the papillae in the young. Thus, the present work aimed to investigate the morphological and structural features of the filiform papillae of neonates.

Materials and methods

Ten specimens were dissected from the anterior part of the tongue from ten stillborn neonates after consent of the parents was obtained. They were then immediately

fixed and processed for both light and scanning electron microscopy.

Light microscopy

Each specimen was sectioned longitudinally into two parts; one part was fixed in formal calcium, washed, dehydrated in ethyl alcohol and embedded in paraffin. Sections of 6 µm were cut and stained with:

- Haematoxylin and eosin
- Periodic acid Schiff for the identification of glycogen after diastase digestion
- Mallory's triple stain to stain collagen fibres and keratin.

Scanning electron microscopy

The remaining parts of the excised tissues were fixed in 4% phosphate-buffered glutaraldehyde for 4 hours at 4 °C, rinsed in buffer and post-fixed in precooled buffered osmium tetroxide. They were then rinsed, dehydrated in ascending grades of acetone and dried using a critical point dryer, They were glued to aluminium stubs, coated with gold and examined with the scanning electron microscope at 15 kV (AMRAY 1200).

Results

Light microscopy

Haematoxylin and eosin (H&E)

Filiform papillae covered the entire anterior part of the dorsal surface of the tongue, appearing as small finger-like projections with rounded or blunted tips; some of them had started to have a conical appearance. In many areas, two or three papillae appeared to originate from one connective tissue base, then cleft formation resulted in splitting of the papillae from each other (Figure 1). Each papilla consisted of a thin parakeratinized epithelial covering and a connective tissue core that showed absence of

secondary papillae and consisted of a fine collagen network, fibroblasts and blood capillaries. The epithelium of the interpapillary regions was nonkeratinized and appeared thicker than that covering the tips and lateral surfaces of the papillae (Figure 2).

Periodic acid Schiff (PAS)

The basal and parabasal cell layers showed mild PAS reaction. The remaining superficial cell layers showed intense PAS reaction in the form of a crescentic deposition of glycogen granules, particularly in the interpapillary regions and at the base of the filiform papillae (Figure 3). The basement membrane and connective tissue showed moderate staining reaction (Figure 3).

Mallory's triple stain

The collagen fibres of the connective tissue were moderately stained (blue colour) and the red blood cells inside the blood vessels appeared red in colour (Figure 4). The thin parakeratin layer covering the filiform papillae was moderately stained (blue colour) (Figure 5).

Scanning electron microscopy (SEM)

Filiform papillae were numerous, regularly distributed over the dorsal surface of the tongue and of different sizes, shapes and at different levels. They mainly appeared as finger-like projections with rounded tips; some of them were conical in shape (Figure 6).

At the tips, the papillae had not yet developed hairs and the surfaces of their bodies were covered by squamous epithelial cells with prominent overlapping cell boundaries. The interpapillary regions contained a network of branching microplicae of the superficial nonkeratinized interpapil-

lary cells. There were no microorganisms covering the body surfaces or the tips of the papillae (Figure 7).

In many areas, filiform papillae appeared lobulated with two (Figures 6 and 8) or three lobes (Figure 9) which originate from one base. This shows the incomplete splitting of individual papillae from each other (Figures 6, 8, 9).

Discussion

The epithelium covering the dorsal surface of the human tongue shows diverse morphological variation from one site to another and even within the epithelium of the same papilla [5]. In the present study, all superficial surfaces of filiform papillae were parakeratinized whereas the epithelium covering the interpapillary regions was nonkeratinized. These results concur with those of El-Zainy [6] and Sawaf et al. [5], and were explained by Dhouailly et al. [7] who suggested that there are different populations of keratinocytes related to different functional requirements, or the different pathways of differentiation of keratinocytes are related to different functional requirements.

Neonates depend mainly on suckling (liquid diet that needs no friction), at least for the first 6 months of life. With the eruption of deciduous teeth and the addition of some types of food to the diet, friction of the tongue occurs in contact with food, teeth and other mucosal surfaces. The parakeratinized epithelium covering the papillae is transformed, orthokeratinized, and the papillae become conlcal in shape. Thus, differences in environment, diet and mastication may modify the morphology and structure of the tongue.



Figure 1 Filiform papillae appear as fingerlike projections with rounded tips; some are conical in shape. Two of them appear to originate from one base (arrows) (H&E × 100)





Figure 2 Two filiform papillae originate from one base (arrow). Each consists of a connective tissue core and a thin parakeratinized epithelial covering. The interpapaillary epithlium is nonkeratinized (H&E × 250)

Figure 3 Filiform papillae showing intense staining reaction in the superficial cell layers, particularly in the interpapillary regions and at the base of the papillae (PAS × 250)



Figure 4 Filiform papillae showing moderate staining reaction in the connective tissue. The three papillae (arrow) start to separate at their tips and the main blood vessel provides branches to the three papillae (Mallory's triple stain × 250)

Figure 5 Moderate staining reaction of the parakeratin layer (arrows) covering the filiform papillae (Mallory's triple stain × 250)





Figure 6 SEM showing filiform papillae (Fi) appearing as finger-like projections with rounded tips. Some appear conical in shape and two of them (arrows) start as one papilla and then split into two papillae (orig. mag. x 180)



Figure 7 Higher magnification of the previous figure showing the surfaces of the papillae covered by squamous epithelial cells (sq) with prominent overlapping raised boundaries (orig. mag. x 500)



Figure 8 SEM showing many filiform (Fi) and fungiform (Fu) papillae and branching microplicae (M) in the interpapillary regions. One filiform papilla (arrow) split again at its tip into two papillae (orig. mag. × 400)



Figure 9 SEM at the tip of the tongue showing three filiform (Fi) papillae with one common base and divided by slight clefts at their tips (arrows) (orig. mag. × 180)

Epithelial mesenchymal interactions play an important role in the development of many organs such as teeth, salivary glands, hair and feathers, but do not occur in the differentiation of filiform papillae. This may simply be due to the relatively small size and close spacing of filiform papillae compared with other epidermal derivatives [8].

A group of unpurified substances known as chalones have been identified in the epithelium and a variety of other tissues. They appear to possess many of the qualities of theoretical morphogens, and appear responsible for the development of filiform papillae [8].

In our study, we observed that the filiform papillae appeared to develop as two or three papillae originating from one common base. Then keratinization and cleft formation occurred which began at the tips of the papillae and progressed downwards until the fused papillae separated from each other. A similar observation was made by Baratz and Farbman [8] during the development of rat filiform papillae. This kind of keratinization and subsequent splitting has been described in the development of several tissues, including the opening of eyelids [9], separation of the digits from each other and separation of the earlobes from the face [10].

The abundance of epithelial glycogen observed in our study in the superficial epithelial layers is in accordance with the observation that glycogen synthesizing enzymes are more prominent in fetal than in adult epithelia [11]. Mcfall and Kraus [12] suggested that glycogen may be present in high concentrations in fetal tissues because of the rapid changes taking place during development which demand quick energy sources.

On examination of sections stained with Mallory's triple stain, the stratum corneum

stained blue confirming the previous findings of Alvares and Meyer [13] and Mc-Millan [14], who observed that, following Mallory's triple stain, the epithelium from different areas stained differently. In some areas the stratum corneum stained yellow or orange and was considered to show complete orthokeratinization, while in other areas the stratum corneum stained blue and was considered to show incomplete orthokeratinization.

The prominent raised boundaries of the superficial squamous cells covering the surfaces of the papillae were similar in appearance to those described by Whittaker and Adams [15] in fetal scalp epithelium and oral mucosa, by McMillan [14] in the hard palate of the rat and by Kullaa-Mikkonen and Sorvari [16] and Kullaa-Mikkonen et al. [2] in human oral mucosa. This arrangement may provide a source of mechanical interlocking in the stratum corneum that resists the shearing forces of mastication [17].

The microplicae of the superficial epithelial cells in the interpapillary regions have been previously described by Cleaton-Jones [17] in human oral mucosa and by Hodgkins et al. [18] in human gingival epithelium. The proposed functional significance of microplicae is for intercellular interdigitation for purposes of adhesion. This serves a protective function by reducing the surface area of contact and aids the flow of surface protecting and lubricating secretions [17, 18].

Conclusion

The filiform papillae of neonates appeared as small finger-like projections with rounded tips, consisting of a connective tissue core and a thin parakeratinized epithelial covering. They appeared to develop as two or three papillae originating from one connective tissue base. Subsequent splitting of the papillae from each other takes place to give their final morphological relationships.

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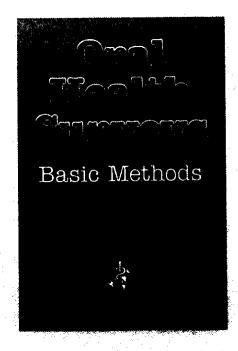
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