S. Afr. J. med. Sci. (1972), 37, 101-104

SURFACE ULTRASTRUCTURE OF THE MUCOSA OF THE SOFT PALATE IN THE VERVET MONKEY

by

P. CLEATON-JONES

Dental Research Unit of the University of the Witwatersrand and South African Medical Research Council, Jan Smuts Avenue, Johannesburg, South Africa.

[RECEIVED 14TH DECEMBER, 1972]

The ultrastructure of the surface of epithelial cells has been described by several workers [Wolf 1967, 1968; Blümcke and Morgenroth 1967; Whittaker and Adams 1971]. All of these showed some form of fine folding on the surface of the cells studied. So constant a finding are these fine surface folds that Marowitz *et al.* [1970] concluded that their presence is a sign of good tissue processing.

Confusion exists however in the interpretation of the appearance of the various types of surface folds. This paper links the two dimensional appearance of the surface folds seen in a transmission electronmicrograph with the three dimensional appearance seen with the scanning electron microscope.

MATERIALS AND METHODS

For transmission electron microscopy (TEM), small pieces of the mucosa of the soft palate were removed from anaesthetised vervet monkeys (*Cercopithecus aethiops*) and fixed in 4.5 per cent phosphate buffered glutaraldehyde solution at pH 7.3 for 1 hour. After washing in phosphate buffer the specimens were post-fixed for 30 min. in 1 per cent phosphate buffered osmium tetroxide at pH 7.3 [Millonig, 1961]. The tissue was again washed in buffer before dehydration in graded ethanol. After the specimens had been passed through propylene oxide, they were embedded in Araldite [Luft, 1961]. Sections were cut on a Reichert OM-U2 ultramicrotome, mounted on copper grids and stained with a saturated solution of uranyl acetate in ethyl alcohol followed by lead citrate [Reynolds, 1963]. The specimens were examined in a Siemens Elmiskop 1 electron microscope.

For scanning electron microscopy (SEM) portions of the soft palates were fixed in a 4.5 per cent glutaraldehyde solution buffered at pH 7.3 with phosphate buffer. After washing in the phosphate buffer the tissue was dehydrated using graded ethanol followed by ether. Alternatively, dehydration was carried out by quenching the tissue in isopentane cooled with liquid nitrogen followed by evaporation under vacuum at —40°C. All specimens were coated with gold palladium and examined in a Cambridge Stereoscan Mark II scanning electron microscope operated at 10kV. The beam/specimen angle was varied so as to obtain the best surface projection.

RESULTS

The cross-sectional view afforded by TEM reveals that the outermost cell surface displays fine projections of fairly constant height separated from each other by a trough also of fairly constant size (Fig. 1). On the surface at the junction between adjacent cells two of the fine elevations lie in close proximity thus giving the impression of a thicker fold on the surface (Fig. 2).

SEM displays a corrugated cell surface; two distinct forms of fine folding can be seen namely (a) fine folds and (b) thicker folds (Fig. 3). The fine folds or microplications are of fairly regular size but with an irregular arrangement. They run in all directions and can show either a linear or a whorled pattern. Whatever the arrangement, the distance between the elevations appears reasonably constant. Many of these fine folds arise independently but a large number are continuous with a thicker

fold that seems to demarcate the boundaries of the individual surface cells. When one of these thicker folds is examined under high magnification a depression is obvious at the crest and the fold can be seen to consist of two parallel microplications (Fig. 4).

If desquamating epithelial cells are also examined by SEM then the undersurface of these cells as well as the upper surface of the underlying cells shows the surface folding (Fig. 5). Similarly if successive layers of cells are examined by TEM then the cellular interdigitations are seen to have the same configuration as the surface elevations (Fig. 6).

DISCUSSION

The finding of fine folding on the surface of cells of the soft palatal epithelium in the vervet monkey is in agreement with that seen in other epithelia by previous workers.

Wolf [1967] using a replica technique described microvilli and bulging ledge-like formations at intercellular borders, which showed an intercellular fissure in their crest. The thicker folds seen in the present study correspond to this description.

Blümcke and Morgenroth [1967] found similar structures in corneal epithelium. Whittaker and Adams [1971], however, in their study of developing skin and oral mucosa, reported intercellular clefts occurring at sites other than on intercellular ridges. Isolated clefts such as they noted were seen in the present study only in a number of the specimens quenched in isopentane cooled with liquid nitrogen. For this reason these clefts are considered to be artefacts.

The microvilli seen by Wolf [1967] were described as fingerlike projections and similar projections were reported in the respiratory tract by Greenwood and Holland [1972]. No such structures were found with the scanning electron microscope in the present study. On the other hand the surface projections seen in the soft palatal epithelium under TEM could quite easily be labelled as microvilli without SEM. These microvilli in a two-dimensional transmission electronmicrograph become cross-sections of the microplications of the surface membrane when SEM findings are also considered.

Elias and Pauly [1966] have drawn attention to the all too common habit of confusing a two-dimensional image with the three dimensional reality. One should therefore exercise care in the use of the term microvillus when applied to two-dimensional electron micrographs. To emphasise this a combination and correlation of the information obtained in the TEM and SEM surface studies is seen in a drawing of the three dimensional appearance of the surface (Fig. 7).

Finally none of the previous authors has described the origin of the microplications. It is felt that they arise due to the exposure of normal cell interdigitations which are uncovered by the exfoliation of surface cells. This is substantiated by the finding of microplications on the under-surface of exfoliating cells as well as the upper surface of the underlying cells. Also the cell interdigitations throughout the epithelial layers have configurations similar to those exposed on the surface.

SUMMARY

The ultrastructure of the surface of the mucosa of the soft palate of the vervet monkey was investigated using both transmission (TEM) and scanning electron microscopy (SEM). The surface of the mucosa shows fine elevations or microplications which have an irregular arrangement except at intercellular boundaries. These elevations are thought to be the remains of normal cell interdigitations.

I wish to thank the Poliomyelitis Research Foundation for making available the monkeys used in this study; the De Beers Diamond Research Laboratories for the use of their scanning electron microscope and Messrs. J. Eckert and R. Greasley for their technical assistance. My thanks also go to Professor D. H. Retief and Drs. W. Evans and J. Austin for their criticism.

REFERENCES

BLUMCKE, S. and MORGENROTH, JR. K. (1967). The stereo ultrastructure of the external and internal surface of the cornea. *J. Ultrastruct. Res.*, **18**, 502-518.

ELIAS, H. and PAULY, J. E. (1966) *Human Microanatomy* 3rd ed., Philadelphia. (F. A. Davis Co., Philadelphia).

GREENWOOD, M. F. and HOLLAND, P. (1972). The mammalian respiratory tract surface. *Lab. Invest.*, **27**, 296-304.

LUFT, J. H. (1961). Improvements in epoxy-resin embedding methods. *J. Biophys. Biochem. Cytol.*, **9**, 409-414.

MAROWITZ, W. F., ARENBERG, I. K. and THALMAN, R. (1970). Evaluation of preparative techniques for the scanning electron microscope. *Laryngoscope*, **80**, 1680-1700.

MILLONIG, G. (1961). Advantages of a phosphate buffer for OsO_4 solutions in fixation. *J. Appl. Phys.*, **32**, 1637.

REYNOLDS, E. S. (1963). The use of lead citrate at high pH as an electron-opaque stain in electron microscopy. *J. Cell. Biol.*, **17**, 208-212.

WHITTAKER, D. K. and ADAMS, D. (1971). The surface layer of human foetal skin and oral mucosa: A study by scanning and transmission electron microscopy. *J. Anat.*, **108**, 453-464.

WOLF, J. (1967). Structure and function of periderm. I. Superficial structure of the peridermal epithelium. *Folia morph.*, **15**, 296-305.

WOLF, J. (1968). Curve of development and disappearance of peridermal phase of human embryonic skin demonstrated by the replica method. *Folia morph.*, **16**, 36-42.

(For legends to plates see next page)

LEGENDS TO PLATES

PLATE 1

Fig. 1. Transmission electron micrograph showing the fine surface projections. × 27,300.
Fig. 2. Cross section of intercellular ridge showing a cleft or depression at the summit. × 54,500
Fig. 3. Scanning electron micrograph showing the surface microplications and the more prominent intercellular ridges. × 3,600.
Fig. 4. High power scanning electron micrograph showing an intercellular ridge consisting of two parallel microplications separated by a cleft. × 45,600.

PLATE 2

Fig. 5. Scanning electron micrograph of an exfoliating surface cell. Note the microplications on both the exfoliating cell above and the underlying cell surface below. × 2,700.
Fig. 6. Transmission electron micrograph of cellular interdigitations approximately midway through the epithelium. × 36,400.
Fig. 7. Diagram of the three dimensional appearance of the microplications and intercellular ridge.