Microvilli of the intestinal mucosal cells of *Rousettus aegyptiacus*

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The microvilli in the small intestine of the bat are very long and slender when compared with those in the rat. This morphology results in the absorption surface per unit area in the bat being three times greater than in the rat. No difference could be observed between the thickness of the plasma membrane of the microvilli and the plasma membrane of the rest of the cell. The terminal web, 'fuzz' and the filaments in the microvilli are all poorly developed in the bat.
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Die mikrovilli van die kleinderm van die vlermuis is baie lank en slank in vergelyking met dié van die rot. Hierdie morfologie veroorsaak dat die absortiewe oppervlakte-area van die vlermuis drie keer meer is dié van die rot. Geen verskille kon waargeneem word in die deursnee van die plasma-membraan van die mikrovilli en die plasma-membraan van die res van die sel nie. Die terminale web, selkleed en filamente van die mikrovilli is swak ontwikkel in die vlermuis.
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The fruit bat, *Rousettus aegyptiacus* has the capacity to assimilate over 16% of its body mass of monosaccharides, taken in as fruit, in a night (Van der Westhuyzen 1974). In a further study Keegan (1977) showed that glucose and fructose were assimilated respectively three and four times faster in the bat than in the laboratory rat yet no evidence for an active transfer of glucose or fructose could be found in the former (Keegan *unpublished*). The small intestine of the bat is only 70% by weight compared to that of the rat.

For these reasons a study was carried out on the ultrastructure of the mucosal cells of the small intestine of the bat to see if the morphology of these cells could shed some light on the mechanisms for assimilation of the monosaccharides. The rat was used as a comparative model.

Methods and Materials

The animals used in this experiment were the fruit bat, *Rousettus aegyptiacus* and the laboratory rat *Rattus norvegicus,* Sprague Dawley strain. The animals were starved overnight of solid food, but had free access to water and an isotonic glucose solution (5,25 g/100 ml). The following morning the animals were killed and the intestine quickly removed and cut into lengths of about 1 cm. These tissues were fixed in a cold (4 °C) solution of a 2% glutaraldehyde in the cacodylate/glucose buffer (pH 7,2 and 415 − 420 mOsm). After an hour the tissues were removed and cut into 1 mm cubes and then returned to the fixative. At the end of four hours the specimens were washed in 0,1 M cacodylate buffer and post-fixed in 1% osmium tetroxide in cacodylate buffer for a further 4 h after which they were washed in the cacodylate buffer and left overnight in this buffer.

The specimens were dehydrated in ethyl alcohol containing 2% uranylacetate and finally placed in propylene oxide. The tissues were embedded in an Araldite/Epon mixture and the thin sections stained with uranylacetate and finally with lead acetate and examined with a JEOL JEM 100C electron microscope.

The measurements of the lengths, diameters and distances between the microvilli were made directly from the negatives. A number of measurements were taken from

each negative and the mean of readings used in the final calculations. The thickness of the plasma membrane was measured both on the negative and the print and here again a number of measurements were made, the mean again being used in the final calculations.

Results

Figures 1 and 2 are typical photomicrographs of the microvilli found in the bat and the rat small intestine cut in longitudinal section. The mean lengths of the microvilli were 3,6 μm and 1,14 μm for the bat and rat respectively, while the average diameters were 0,099 μm and 0,14 μm (Table 1). Figures 3 and 4 show the microvilli cut in cross-section so that the distance between the microvilli as well as the diameters could be measured. In the bat and the rat the distance between the microvilli was 0,043 μm and 0,038 μm respectively.

The cross-section photographs showed that there was a roughly hexagonal arrangement between the microvilli and that there were approximately 57 and 36 microvilli per μm^2

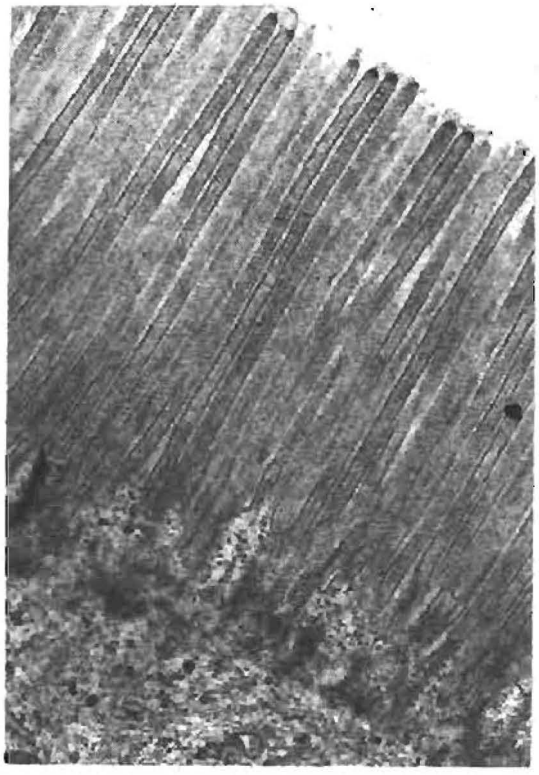

Fig. 1 Longitudinal section of the microvilli in the bat's jejunum × 20 000.

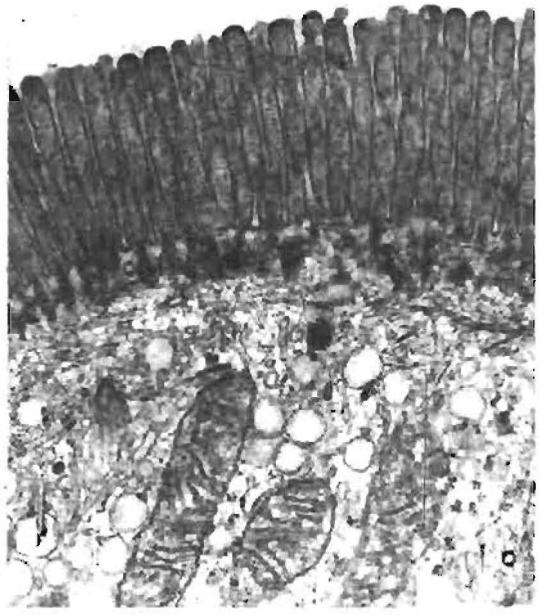

Fig. 2 Longitudinal section of the microvilli in the rat's jejunum × 20 000.

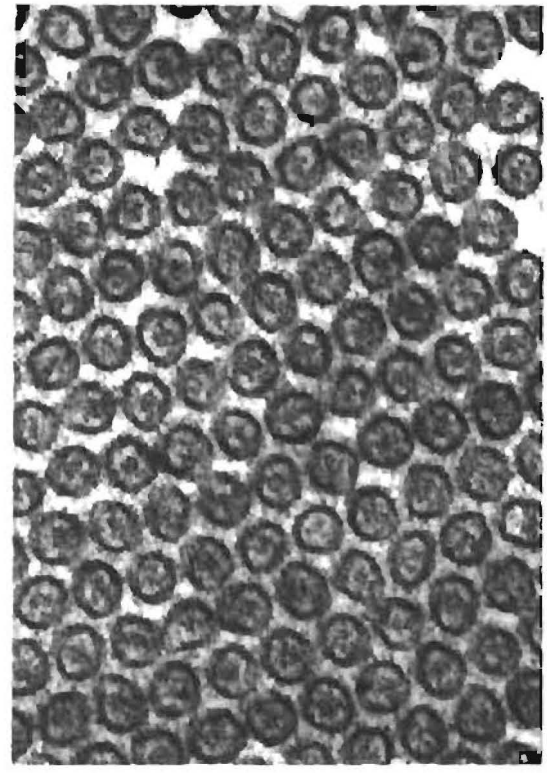

Fig. 3 Cross section of the microvilli in the bat's jejunum × 60 000.

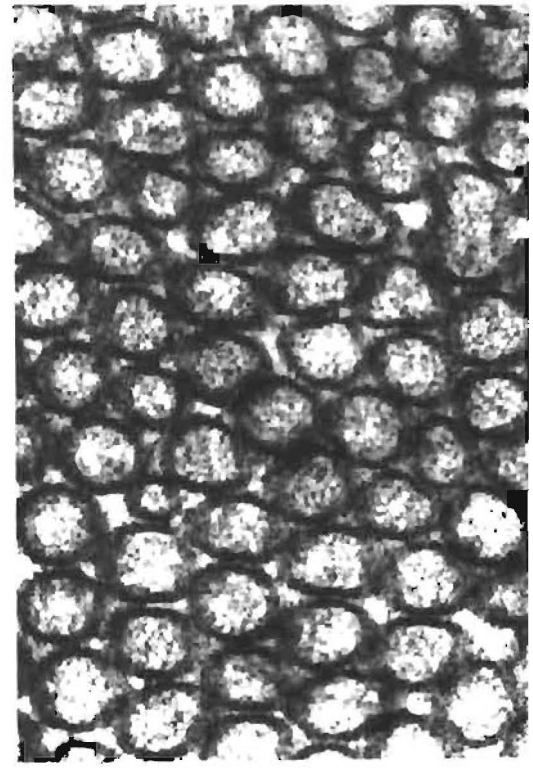

Fig. 4 Cross section of the microvilli in the rat's jejunum × 60 000. (The 'Fuzz effect' on the photograph tends to obscure the true width of the plasma membrane).

Table 1 Summary of the morphology and the surface areas of the microvilli in the bat and the rat

	Bat	Rat
Length of microvilli (μm)	3,6 (15)*	1,14 (10)*
	S.D. ± 1,0	S.D. ± 0,2
Diameter of microvilli (μm)	0,099 (22)	0,14 (7)
	S.D. ± 0,01	S.D. ± 0,01
Distance between microvilli (μm)	0,043 (22)	0,038 (5)
	S.D. ± 0,005	S.D. ± 0,001
Surface area of microvilli (μm^2)	1,0	0,5
No. of microvilli/μm^2 calculated	57	36
No. of microvilli/μm^2 counted	56,5 (9)	35,2 (5)
	S.D. ± 2,7	S.D. ± 3,7
Therefore surface area increase due to microvilli	57	18

*Number of observations in brackets.

Table 2 The thickness of the plasma membrane

	Bat	Rat
Microvilli	80,7 Å (19)*	73,0 Å (4)
	S.D. ± 9,7 Å	S.D. ± 17,0
Lateral membrane	81,0 Å (7)	74,0 (4)
	S.D. ± 10,6 Å	S.D. ± 18,1
Basal membrane	86,0 Å (11)	73,0 (4)
	S.D. ± 10,6 Å	S.D. ± 16,7

*Number of observations in brackets.

in the bat and rat respectively.

In the bat the thickness of the plasma membrane of the microvilli was of the same order as that of the plasma membrane on the lateral and basal aspects of the cell. Similar results were observed in the rat. The thicknesses of the plasma membranes are given in Table 2.

Discussion

Examination of the gross anatomical and light microscopic structures of the small intestine of the bat and the rat shows that they are similar. Therefore it would appear that the gross structure of the bat's small intestine does not explain the very rapid rate of glucose absorption as seen in the bat.

The most striking feature when comparing the ultrastructure of the intestinal mucosal cell in the bat and the rat are the microvilli. The microvilli in the bat are long and slender with an average length and diameter of 3,6 μm and 0,1 μm compared to the rat's 1,14 μm and 0,14 μm. This length of 1,14 μm for the rat's microvilli, compares well with that of 1 μm reported by Millington and Finean (1962), and Palay and Karlin (1959) for the rat. Thus the bat's microvilli are over three times longer than the rat's. In other reported species the value varies from 0,7 – 1,9 μm Merrill *et al.* (1976) and Shearman *et al.* (1962).

Unlike Brown (1962) and Merrill *et al.* (1976) we could show no significant difference between the lengths of the microvilli with respect to the situation of the cell as to its position on the villus, nor was there any significant difference between the lengths of the microvilli found in the duodenum, jejunum and ileum. There was, however, a tendency for the longest microvilli to be those at the crest of the villi in the distal part of the jejunum in the bat.

The diameter of the microvilli in the bat was less than in the rat, 0,099 μm cf 0,14 μm. The diameter of the microvilli in the rat is larger than previously reported, where the general finding has been that it is approximately 0,1 μm, Millington and Finean (1962), and Palay and Karlin (1959).

In Figs. 3 and 4 the arrangement of the microvilli appears to be a hexagonal pattern and the distance between the microvilli in the bat was 0,043 μm and 0,038 μm in the rat. As the length, diameter and distance between the microvilli were known, the total surface area of an individual microvillus and its supporting plasma membrane could be calculated. In the bat this area was found to be approximately 1,0 μm^2 whilst in the rat approximately 0,5 μm^2. From the theoretical area of the hexagon supporting the entire microvilli the number of microvilli per μm^2 of the theoretical flat apical surface of the mucosal cell could be calculated. The number of microvilli per μm^2 was calculated to be approximately 57 in the bat and 36 in the rat. The calculated figures for the number of microvilli per μm^2 in both the rat and the bat were the same as the number of microvilli counted in the cross-sectional specimens (Table 1). As the calculated and observed numbers of microvilli per unit are so similar, the assumption that the microvilli were arranged in a hexagonal pattern is probably correct.

Because the surface area of the microvilli units, and the number of microvilli per μm^2 were known, the total surface area of the microvilli and supporting plasma membrane per μm^2 of the theoretical apical surface area could be calculated. The results showed that due to the microvillus structure there was a 57 and 18 fold increase in the mucosal surface of the intestinal cell in the bat and the rat respectively. Palay and Karlin (1959) reported a 24 fold increase in the rat. Thus in the bat the assimilation surface of the small intestine is three times greater than in the rat. This means that if this plasma membrane was the limiting factor with regard to the rate of absorption then the very rapid rate of absorption of the monosaccharide observed in the bat could be accounted for in part by this increase in the surface area.

Another interesting observation is that the distance between the microvilli in the bat is greater than in the rat. This greater distance between the microvilli should mean that in the bat there is a better access for the intestinal contents to the basal regions of the microvilli.

No significant difference was observed between the thickness of the plasma membrane found in the microvilli or the lateral and basal aspects of the cell in either the bat or the rat (Table 2). This was contrary to what had generally been found in other species including the rat where the plasma membrane of the microvilli was thicker, 95 to 115 Å, compared with 70 to 80 Å for the rest of the cell. Sjöstrand (1963) reported a value of 95 Å for the plasma membrane of the brush border in the mouse and suggested that a possible reason for his lower plasma membrane figure was the higher resolution and the thinner sections he used compared to previous workers. Suganuma (1961) and Merrill *et al.* (1976), however, reported a figure of 70 Å for the plasma membrane of the microvilli in both the frog and the guineapig.

The structure of the plasma membrane in the bat varied considerably, even in the same section, for in some areas the membrane was symmetrical while in others asymmetrical. The filaments in the microvilli were not prominent when the microvilli were sectioned along their lengths but were observed when the microvilli were cut in cross-section although they do not appear as abundant as in the rat. The terminal web likewise in the bat is poorly developed when compared with other species. Mooseker and Kilney (1975) suggested that this complex of filaments and terminal web was responsible for the movement of the microvilli. As the bat assimilates monosaccharides many times faster than the rat, then it would appear unlikely that movement of the microvilli is associated with the assimilation of those sugars.

The fuzz on the microvilli, suggested by Hamilton and McMichael (1968) and Pritchard (1969) to act as a possible barrier to the movement of glucose back into the intestinal lumen after the hydrolyses of maltose, is poorly developed in the bat. However, it would appear that this structure is not important in the bat for the rate at which this species assimilates the products of the hydrolysis of sucrose, is of the same order as when a mixture of glucose and fructose of equivalent concentrations are given (Keegan *unpublished*).

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