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Microanatomy of the Fishing Bat Skin

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Abstract.- In this study, we provide a comparison of the epithelial histology of fishing bats (*Genus species*) in different body locations (dorsum, abdomen, lip, intercrural membrane, wing, footpad and head). We found two main types of external epithelia - glabrous skin and hair-bearing skin. The skin of this species consisted of three components: the epidermis, the dermis and hypodermis, with an average thickness of 308 μm. The dorsal skin was the thickest (605 μm). In most locations of the bat's body, the epithelial layer was composed of one to three cell layers, except in the thick skin of the footpad, which had more than 3 layers. Among all the body parts, the footpad had the thickest stratum corneum (31±2 μm; Mean ± SD) and epithelial layer (35±7 μm). The glabrous skin had no sebaceous glands. The thickness of the epithelial layer was positively related with the thickness of the stratum corneum but negatively related with the density of hair coat.

Keywords: skin, stratum corneum, hair follicles, histology, *Myotis ricketti*

INTRODUCTION

Bats are one of the most extraordinary mammal orders because of their adaptations for flight. The fishing bat was first described by Thomas (1894). In China, the fishing bat (*Myotis ricketti*) was first described in Beijing in 2003 (Ma *et al*., 2003b) and subsequently a large population has been found in Beijing (Ma *et al*., 2003a).

The skin is a complex organ covering the whole surface of the body. There are many studies about skin histology in different mammal species (Laruche and Cesarini, 1992, Barbara *et al*., 1997; Kanitakis, 2002; Thomas, 2005). In contrast to the exhaustive research focused on the skin of other mammal species, limited information is available about the structure of bat skin. Existing information is either restricted to limited areas (Quay, 1969) or only gives a simple introduction to the structure of the skin (Quay, 1970). Understanding the structure of bat skin can offer the basis for morphofunctional studies in these specialized mammals.

Generally, the skin of mammals can be divided into three components: epidermis, dermis, and hypodermis. The epidermis is made up of two layers, namely epithelial and stratum corneum. The epithelial layer is largely composed of stratified squamous epithelium and a variable amount of connective tissue containing melanocytes, Langerhan′s cells and Merkel cells. The stratum corneum is a protective layer consisting of keratin-impregnated cells. At the same time, the satratum corneum usually varies considerably in thickness and has a typical basketweave appearance. The dermis, containing sensors, receptors, blood vessels and nerve ends, consists of two structurally different layers named papillary and reticular. The hypodermis is composed of loose connective tissue and fat.

The main objectives of this study were (1) to examine and describe more fully the microanatomy of normal bat skin, and (2) to compare the skin structure in different parts.

MATERIALS AND METHODS

Bat material

All procedures involving animals were carried out in accordance with the Policy on the 106 Care and Use of Animals, approved by the Ethical Committee, Division of Animal Ecology and Conservation Biology, Institute of Zoology, Chinese Academy of Sciences. Five fishing bats were collected using mist nets in a large cave located in Xiayun Ling, Fangshan District, Beijing (115° 59' N, 39° 43' E).

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Skin preparation

The bats were killed by decapitation. To allow observations of the surface of the skin, depilation cream (Shibi, Beijing, China) was used to remove hair. Skin samples (2 cm^2) were taken from the footpad, wing, dorsum, abdomen, head and intercrural membrane and immersed overnight in 4% paraformaldehyde in 0.06 M phosphate buffer (pH 7.3) solution.

Histological study

Skin samples were dehydrated through a graded series of ethanol, cleared in dioxane, and embedded in paraffin. Four-micron-thick serial sections were deparaffinized and stained with Harris's hematoxylin and eosin. Finally, they were examined under a Nikon (AXIO-OEVT-35M, German) light microscope and photos taken. Ten fields of view were chosen for per body region. The thickness of the skin and its component layers (epithelial layer, stratum corneum, dermis and hypodermis) was measured using a computerized light microscope (Nikon, AXIO-OEVT-35M, German) and spot basic software (USA).

RESULTS

The skin of bat is divided into three layers, the epidermis (epithelial layer and stratum corneum), dermis and hypodermis. The microanatomical features of the skin from each part of the bat body are as follows:

Dorsum (Figs. 1A, B)

The average skin thickness, from the top of the stratum corneum to the bottom of the hypodermis, was 605±22 μm. The thickness of stratum corneum was 19±1 μm. The epithelial layer consisted of 1 or 2 cell layers with an average thickness of 15±1 μm. The dermis was divided into two parts, papillary and reticular. The average thickness of the dermis was 168±10 μm. The reticular dermis contained many collagen fibers, with abundant sebaceous glands, hair follicles and a few merocrine sweat glands immersed. The dermis and hypodermis were easily distinguishable and the latter include scanty apocrine sweat glands.

Many hair follicles of fishing bat were associated with sebaceous glands (Figure 1A, B). The hair follicle was made up of three parts: the lower portion, which extends from the base of the follicle, including hair bulb, to the insertion of the bulge; the isthmus that projects from the insertion of the bulge to the entrance of the sebaceous duct; the infundibulum, which comprised the entrance of the sebaceous duct to the follicular orifice.

Abdomen (Figs. 1C, D)

The mean skin thickness was 319±11 μm. The stratum corneum was 13±1 μm thick. The epithelium consisted only in one cell layer, with the thickness of 12±1 μm. The dermis (66±8 μm) had abundant sebaceous glands and there were plenty of exocrine sweat glands. Only loose connective tissue was found in the hypodermis.

Lip (Figs. 1E, F)

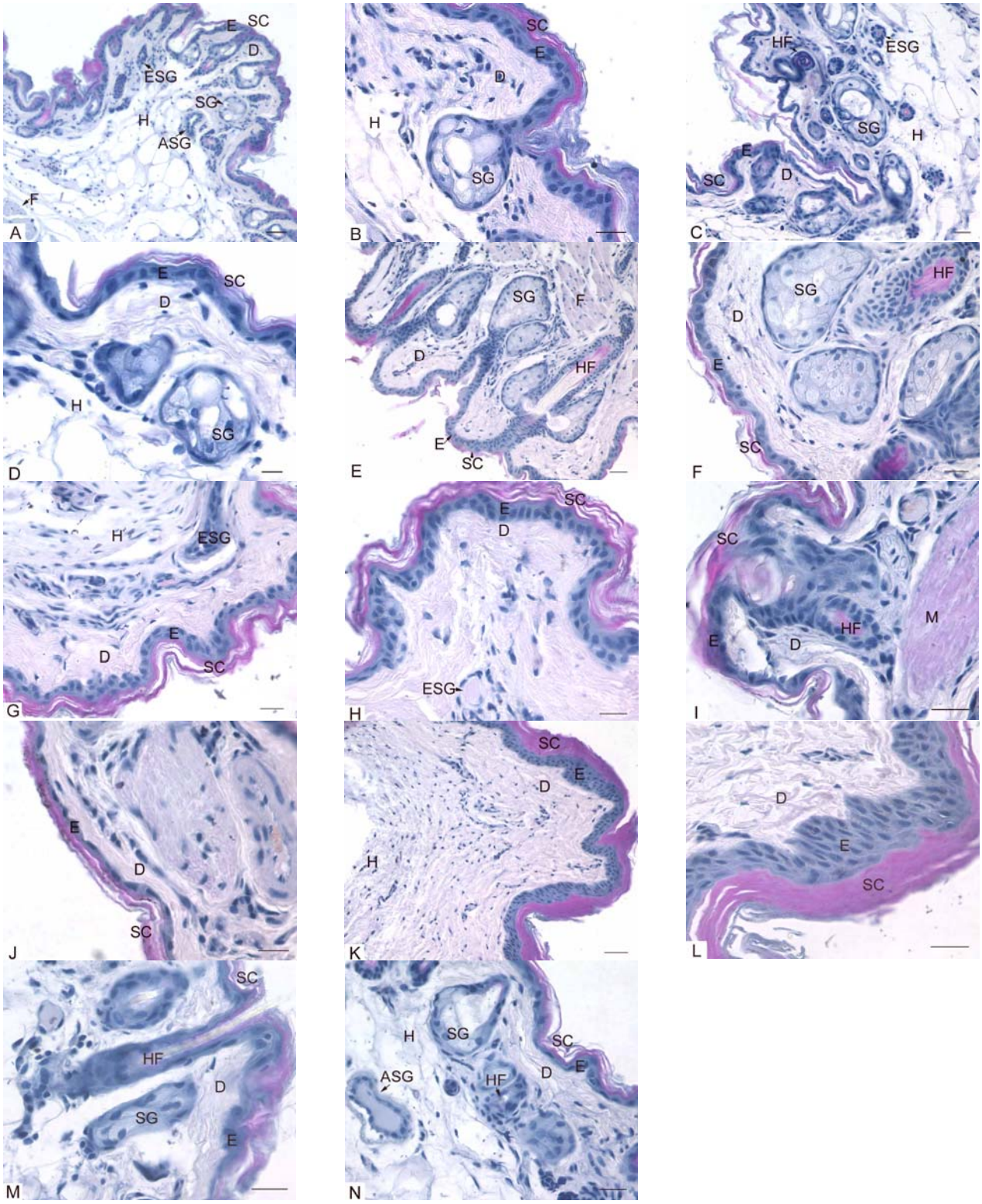
The average skin thickness of the lip was 336 ± 9 μm. The stratum corneum was 7±1 μm thick. The epithelium (13 ± 2 μm) was monolayered. The dermis had an average thickness of 139±9 μm and contained a large amount of sebaceous glands than elsewhere of the fishing bat. The hypodermis was composed of loose connective tissue and fat.

Intercrural membrane (Figs. 1G, H)

The average skin thickness of the intercrural membrane was 148±9 μm. The stratum corneum was 22±1 μm thick. The epithelial layer (20±3 μm) contained two layers. Within the dermis (91±5 μm), there were less sebaceous glands but more collagen fibers than dorsum. The hypodermis consisted of connective tissue.

Wing (Figs. 1I, J)

The wing had many domes with a hair emerging from each one. The average skin thickness was 52±6 μm, of which 11±2 μm was comprised by the stratum corneum. The epithelial layer was 9±2 μm thick. The wing was divided into two parts, the plagiopatagium and the wing membrane. The array of cells was sparser in wing membrane than in plagiopatagium. Few sebaceous glands layed in the dermis. Within the hypodermis, the collagen fibers were larger than elsewhere in the fishing bat's body and were arranged into bundles.



For legend, see page 390

Footpad (Figs. 1K, L)

The average skin thickness was 349±14 μm. The stratum corneum was the thickest layer of all the body regions, 35±7 μm. The epithelial stratum consisted of 3-6 cell layers, and its thickness was 31±2 μm. There were no sebaceous and sweat glands in the dermis, whose average thickness was 192±14 μm. The dermis and hypodermis consisted of collagenous fibers.

Head (Figs. 1M, N)

The average skin thickness was 337±11 μm. The stratum corneum was 11±2 μm thick. The epithelium, whose average thickness was 15±2 μm, consisted of one cell layer. The cells in epithelium were scatteredly arranged. Many hair follicles extended through the dermis. Sebaceous glands were found on both sides of the hair. The average thickness of the dermis was 124±7 μm.

Comparisons among different parts of skin

The skin of the dorsum was the thickest, having both the thickest dermis and hypodermis. The stratum corneum and the epithelial layer of footpad was the thickest. The skin of the wing was the thinnest.

DISCUSSION

Classification of bat skin

There were two main kinds of bat skin, glabrous skin (found only on the footpad) and hair-bearing skin. The glabrous skin was characterized by a thick epidermis. The hair-bearing skin held many follicles and sebaceous glands. Both characteristics are similar to some mammals (Ferry *et al*., 1995; Barbara *et al*., 1997; McGrath, 2004).

The thickness of the skin

In the fishing bat, the epithelial layer of most body parts was made of 1-3 cell layers. The skin of the dorsum was the thickest whereas the skin of the wing was the thinnest. Moreover, the dermis of the dorsum was the thickest part of the entire dorsal skin. These results agree with the conclusion that the dermis is the main component contributing to skin thickness in most mammals (Scott, 1988).

As for why the thickest skin is located on the dorsum rather than other parts of the bat body, it may be because there are many hairs in this area, and the dermis in the dorsum can offer space for the hair. In addition, thick skin can protect the bat from the bad weather and prevent them from being attacked

Stratum corneum

Cells in stratum corneum were highly flattened because they have lost nuclei and cytoplasmic organelles. The keratin filaments align into disulphide cross-linked macrofibres (McGrath, 2004). The stratum corneum of haired skin tended to be thin, whereas it was thicker in areas of non hairy skin coat. The stratum corneum of the footpad in fishing bat was the thickest. This may be associated with the fact that bats use the footpad to grasp an object. The situation was supported by the study about humans (McGrath, 2004) and domestic carnivores (Banks, 1981). The larger number of cell layers and thicker stratum corneum have great implications in maintaining life as the barrier of the

Fig. 1. Skin histology from different body locations with Harris's hematoxylin and eosin staining. (A) Transverse section of the skin of the dorsum. The skin is composed of three parts, epidermis (E), dermis (D) and hypodermis (H). Scale bar = 40 μm. (B) Showing 1-2 cell layers of epithelial layer (E) and sweat glands in dermis of the dorsum. Scale bar = 10 μm. (C) The epidermical cells, eccrine sweat glands (ESG), sebaceous glands (SG) and hair follicles (HF) of the abdomen. Scale bar = 20 μm. (D) The sebaceous glands of the abdomen. They lie mainly in the dermis. Scale bar = 20 μm. (E) Showing hair follicles and a mount of sweat glands of the lip. Sweat glands mainly lie around the hair follicle. Scale bar = 30 μm. (F) Sweat glands of the lip in detail. Scale bar = 20 μm. (G) Four layers of the intercrural membrane skin. Scale bar = 20 μm. (H) Eccrine sweat glands of the intercrural membrane. Scale bar = 20 μm. (I) Dome and muscle of the wing. Scale bar = 20 μm. (J) Detail of the arrangement of the cells in the wing membrane. Scale bar = 15 μm. (K) Skin of the footpad showing all parts. The epithelial layer is made up of several cell layers. Scale bar = 40 μm. (L) The stratum corneum (SC) of the footpad skin. Scale bar = 15 μm. (M) The hair follicles of the head. Scale bar = 20 μm. (N) The sweat glands and apocrine sweat gland (ASG) of the head's skin. Scale bar = 20 μm.

skin. Additionally, the direct proportionality between the thickness of the stratum corneum and the thickness of the epithelial layer and the inverse relationship between the thickness of stratum corneum and the density of the hair coat, seems to follow the same patterns as found on the skin of llamas (Barbara *et al*., 1997).

Hair follicles

Our study showed that hair follicles of the body were present in a relatively high density, and were orientated at a sharp oblique angle. This arrangement is suspected to provide insulation and protection from unfavorable environments. The marked angulation of the hair follicles seen in bat skin has also been reported to be a feature of llama (Barbara *et al*., 1997) and is contrast to sheep, in which the follicles lie almost perpendicular to the skin surface (Dowling and Nag, 1962). Simple hair follicles are associated with sebaceous glands and a sweat gland (Spearman, 1977). The fishing bat has simple hair follicles, which is the same as in horses, cattle and pigs (Blackburn, 1965).

Sweat glands and sebaceous glands

There were a few sebaceous glands and sweat glands in the skin of all the body parts examined in this study, except for the footpad. In the fishing bat, there were no sebaceous glands and sweat glands in footpad. This is different from the situation in human, which has 300 glands/cm^2 throughout the footpad (Kanitakis, 2002). Sweat glands have been suggested to be important for evaporative cooling (Rosychuk, 1989). Results from this study indicate that the footpad of the fishing bats may not have the function of evaporative cooling.

CONCLUSIONS

Although the basic histological components of bat skin were found to be similar to some mammalian species, certain specialties exist in the bat's skin. First, the arrangement of the cells becomes sparse from plagiopatagium to wing membrane. Second, in most parts of the bats body, the epithelial layer is composed of one to three cell layers, except on the footpad. Several questions should be answered by further studies, such as why the skin of dorsum is the thickest among all body parts and the reason for the arrangement patterns of the cells in the wing.

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