Influence of Rain on Activity of the Indian Flying Fox
Pteropus giganteus

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ABSTRACT

Because individuals of colonies of the Indian flying fox, Pteropus giganteus openly roost in trees by exposing themselves to broad daylight, we raised a question on their adaptation to rains. We arbitrarily divided the rains into three categories light, medium and heavy. When rain occurred during daytime majority of the bats wrapped the ventral parts of their bodies including heads and remained motionless. Although emergence flights occurred when rains occurred during evenings, relatively less number of bats emerged even in heavy rains when compared to the other two kinds of rains. It appears that bats were thirsty as they began to move around soon after cessation of the rains and lapped on the drops of water that remained on the leaves. Histological study indicates that the dermis layers of wing membranes contain sebaceous glands with ducts in P. giganteus and without ducts in other bats such as Cynopterus sphinx, Hipposideros speoris and Megaderma lyra. Fluorescent microscopic study showed the presence of water proof substance, sebum in the glands. Presumably sebum may be produced to reach the upper regions of wing membranes of P. giganteus, to form a protective layer. SEM analyses showed that the wing membranes of P. giganteus are significantly thicker than those of the other bats.

Keywords: Rain, Sebaceous gland, Pteropus giganteus, Cynopterus sphinx, Hipposideros speoris, Megaderma lyra

Abbreviations: SEM – Scanning Electron Microscope.
1. INTRODUCTION

Nearly all species of bat (except species of the genus *Pteropus*) occupy a wide variety of habitats like caves, crevices, temples, ruined buildings and foliage. Bats living in such habitats are well protected from predators and the abiotic factors like sunlight, ambient temperature, humidity, thunder and rain. Whereas, bats belonging to the genus *Pteropus* live in trees by exposing themselves to these abiotic factors. *P. giganteus* is one among the 13 pteropodids that are distributed throughout India (Bates and Harrison, 1997). It lives in colonies consisting of hundreds in trees like banyan, tamarind, mast and eucalyptus. During hot summer days majority of the individuals gently flap the distal end of one of their wings, which is attributed to thermoregulation (Nelson, 1965; Jones, 1972; Mathur et al., 2012). When it rains during daytime, individuals of *P. giganteus* wrap their bodies including heads and remain motionless till cessation of rain. Similar to gentle wing flapping exhibited during hot days, *P. giganteus* may possibly be equipped with a system to alleviate wetting their bodies by rain. Because bats cover their bodies with wing membranes while raining, we predicted that oil glands may prominently be found in the wings compared to that of other species of bats that live in protected habitats like caves. We carried out a study on a colony of *P. giganteus* by observing the behaviour exhibited by the individuals during and after rains during daytime and emergence flights during evening hours. In addition, we carried out histological study on the ultra structure of wing membranes of *P. giganteus*, *Cynopterus sphinx*, *Meadorma lyra* and *Hipposideros speoris*.

Categories of rain:
We arbitrarily divided the rain into three categories based on the need for an umbrella. ‘Light’ rain in which umbrella is not needed. We need umbrella for the other two categories.

Videography:
After the cessation of rain during daytime, the bats started moving around the branches and licked the water droplets that remained on leaves. This behaviour was videographed.

Scanning Electron Microscopy:
SEM analysis was used to measure the thickness of the wing membranes of four species of bat.

Fluorescent Microscopy:
In order to identify the presence of sebum producing sebaceous glands this method was used.

2. MATERIALS AND METHODS

The study was carried out from October 2009 to December 2010 on a colony consisting of about 1000 individuals of *P. giganteus*. The colony occupied various branches of an *Albizia lebbeck* and three *Ficus benghalensis* trees, which were located opposite to the Vice-Chancellor’s residence inside the Madurai Kamaraj University campus. Visual observations with the aid of binoculars (Balileo, 20x50 Grossfeld, 8m/180000m) were made during daytime on rainy days that provided a total of 673 observation hours. Observations were continued for one hour after the cessation of rains. During evening hours of a total of 25 days, we observed the timings of onset and number of individuals that departed the trees during their emergence activity. In addition, we visually spotted the flying bats after their departure by stationing ourselves at four different places, two sites each at eastern and western sides that were approximately at a distance of 3 km to each other to a total of 6 km away from the day roost tree. Based on the necessity of umbrella for us, we arbitrarily divided the rain into three categories – light, medium and heavy. We did not need umbrella when the rains were light. Whereas, umbrella was necessary for the other two kinds of rains. The ‘intensity’ of the rain was relatively higher during heavy when compared to medium rain.

Because bats wrapped ventral part of their bodies with wing membranes when raining, we carried out histological study on the structure of the wings. In order to compare the structure of the wing membrane of *P. giganteus* with other species, the histological study was done on another pteropodid bat, *C. sphinx* and two microchiropterans namely *M. lyra* and *H. speoris*. Wing samples were collected with 3 mm biopsy punches from the plagiopetagium. The samples were fixed with 2.5% glutaraldehyde and 2% paraformaldehyde, washed in 0.1M phosphate buffer and post fixed with 1% osmium tetroxide. After dehydration with 70%, 90%, 95% and 100% ethanol, the samples were dried by using critical point dryer (Emitech K850, England). Afterwards, the samples were mounted on metal pedestals, coated with gold palladium alloy (JEOL JFC 160 Auto fine coater, Japan) and examined under scanning electron microscope (JSM-6490LV, JEOL, Japan) at different magnifications. The images were photographed.

3. RESULTS

Rains occurred at various timings from 10.00 to 19.20 h for a total of 25 days during the study period. It was possible that the same rain changed to other categories, i.e. light to medium and then to heavy on the same spell. The duration of raining ranged from 5 to 145 min. When raining, all individuals of the colony of *P. giganteus* wrapped ventral part of their bodies with wing membranes and hung silently without showing any movement. About 10 min after the rain stopped, the bats fluttered their wings, groomed the bodies with claws of hind limbs and licked thoroughly the inner and outer areas of wings for 9.1±0.64 sec (n=22), 56.0±5.95 sec (n=22) and 13.3±0.64 min (n=22) during the respective three rains. Afterwards, each individual began to move around the branches of the trees and licked (n=187) the water droplets that remained on the leaves by swiftly stretching their tongues (S1). An individual *P. giganteus* covered approximately 30 leaves in a minute on such activity (See Video S1).

During evening hours we observed a total of 2162 individuals of *P. giganteus* exhibited emergence flights in 25 days, 386 in 10 days and 184 in 8 days during light, moderate and heavy rains, respectively (Fig. 1). We observed a total of 51 bats in heavy rains crossing a place known as Kokkulam that was at a distance of 6 km away from the day roost.

Histological study showed that the wing membranes of *P. giganteus*, *C. sphinx*, *M. lyra* and *H. speoris* were composed of three layers – epidermis, dermis and hypodermis. The epidermis was covered by stratum corneum. The dermis layer contained sebaceous glands, sweat
Since individuals of P. giganteus are directly exposed to hot and cold environments including rain, a comparative study on their wing membranes with other species of bats that live in well protected habitats was expected to provide new scientific information.

4. DISCUSSION
The present study showed that individuals of P. giganteus adapt themselves not only to the simmering hot temperatures during summers (~42°C in Madurai and even more in northern regions of India), but also when exposed to heavy rains.

Based on our prediction, the histology study revealed the presence of oily substance-producing sebaceous glands in the wing membranes in P. giganteus. The presence of duct in the gland only in P. giganteus suggests secretion of sebum that may presumably be followed by occurrence of hydrophobic reactions to withstand the wet. Since the bats began to move around the branches of the trees soon after the rain stopped, their immobile posture when raining may not be attributed to torpor. Drinking of water soon after the rains stopped suggests that the bats were thirsty.

Heavy rains delay emerging time in insectivorous bats that leads to reduced activity (Audet, 1990; Catto et al., 1996). Rain interferes with echolocation or negatively affects thermoregulation (Fenton et al., 1977). In addition, Christian et al. (2011) have reported that rain increases the energy cost of bat flight. The feeding period of fish-eating bat, Noctilio leporinus is shifted to later time during rain (Bork, 2006). The foraging activity of Carollia castanea is not interrupted by light and moderate rains, but stopped, by heavy rain (Thies et al., 2006). The foraging activity of C. sphinx is not affected by rain without lightning and thunder whereas during rains with thunderstorms, the average foraging time was delayed (Yu et al., 2010). The emergence activity of megachiropterans, especially P. giganteus, is apparently not affected by rains.

The pilosebaceous unit in the bat wing is considerable potential interest as a model for fundamental study of sebum production (Cortese and Nicoll, 1969). The waterproof agent, sebum, protects the human and other mammals’ skin and hair (Strauss et al., 1991; Thiboutot, 2004). In birds preen gland releases the sebum that facilitates keeping their feathers water resistant (Elder, 1954). The fishing bat Myotis ricketti possesses sebaceous gland, which is located in dermis layer of wing membrane and secretes sebum to lubricate and make it waterproof (Yin et al., 2011). Two types of sebaceous gland (association with hair follicles and free glands) are present in the dermis layer of P. giganteus, whereas C. sphinx has free sebaceous glands. Wing membranes of the fruit bat Epomophorus wahlbergi has no glands. In the microchiropteran bat H. speoris less number of free sebaceous glands are found in its wing samples. In contrast, Indian false vampire bat M. lyra has not possessed any glands in its wing membrane. The relatively thicker wing membrane and the presence of more sebaceous glands might be an adaptive strategy evolved in P. giganteus, to tolerate hot and wet conditions.

5. CONCLUSION
Among the 1200 species of bats that are distributed throughout the world, only the Pteropus species live mainly on trees exposing themselves to the daylight. All other species live in closed areas such as caves, crevices, unused buildings etc. Hence individuals of Pteropus must adapt themselves to hot and cold temperatures as well as rain. We arbitrarily divided the rain into light, medium and heavy. Emergence flights during evening hours were not completely stopped even during heavy rain. When it was raining, individuals of P. giganteus wrapped their entire bodies and heads with their wing membranes. Histological study showed that the dermis layer of the wings contained sebaceous glands that secrete oil like substance known as sebum. The glands also contained duct that open into hair follicles. Hence it is possible that when raining occurs, sebum may be produced and protects the wing membranes. Interestingly although other species of bats (Cynopterus sphinx, Hipposideros speoris and Megaderma lyra) contained sebaceous glands, the duct is not found in their wing membranes. In addition, SEM analyses showed that the wing membranes of P. giganteus are relatively thicker than those of other bat species.

SUMMARY OF RESEARCH

1. Majority of the bat species live in protected areas like caves, crevices, abandoned buildings, tree holes etc. Whereas individuals of Pteropus species live in trees and are exposed to direct daylight.
2. We undertook a study on a colony of P. giganteus to find out how the individuals adapt themselves to rain.
3. When rain occurred during daytime, all the individuals wrapped their bodies and heads with their wing membranes. Histological study showed that sebum producing sebaceous glands with ducts opening into hair follicles are found in the dermis layer of the wings of P. giganteus. Although sebaceous glands are present in other species of bat such Cynopterus sphinx, Hipposideros speoris and Megaderma lyra, their glands were ductless.
4. SEM analysis showed that the wing membrane of P. giganteus is relatively thicker than that of other species.
5. In spite of heavy rain, emergence flights during evening hours were not completely stopped.
FUTURE ISSUES
The sebum component that was found in the sebaceous gland is to be further taken into biochemical analysis.

DISCLOSURE STATEMENT
There is no special financial support exclusively for this work from the funding agency.

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REFERENCES
Figure 1
Three categories of rainfall and the number of bats emerged during evening hours during such rains over the seasons

Table 1
Thickness of wing membranes of Mega and Microchiroptera

<table>
<thead>
<tr>
<th>No.</th>
<th>Wing thickness (µm)</th>
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<tr>
<td></td>
<td>P. giganteus</td>
</tr>
<tr>
<td>1</td>
<td>223.33</td>
</tr>
<tr>
<td>2</td>
<td>228.78</td>
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<tr>
<td>3</td>
<td>253.29</td>
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<td>4</td>
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<td>5</td>
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<td>6</td>
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<td>SE</td>
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http://www.discovery.org.in/ijss.htm
Figure 2

Figure 3
Figure 2
Histological structure of sebaceous glands in the wing membrane of Mega and Microchiroptera

Figure 3
Fluorescent microscopic view of water proof agent “sebum” containing sebaceous glands on wing membrane of of Mega and Microchiroptera;
(a - P. giganteus; b - C. sphinx; c - M. lyra d - H. speoris, SG: Sebaceous gland; D: Dermis; ED: Epidermis; HD: Hypodermis; HF: Hair follicle)

S1: Video of P. giganteus moving around branches of tree and licking the drops of water after the rains stopped during daytime (See video in Online)