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Original article

Histomorphometry of the lower respiratory system of straw coloured fruit bat (eidolon helvum)

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ABSTRACT

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The description of microanatomy of the lower respiratory tract (LRT) in the straw coloured fruit bat is scarce. Therefore, the present study is planned at documenting normal histology of the LRT of the bat (*E. helvum*) found in Nigeria. The tracheal, principal bronchi and lungs were cut at 5 μ m thickness sectioned for histomorphology. Tissues from seven (7) bats were utilized in the study. Sections of the LRT were stained with Haematoxylin and Eosin (H & E) stain. The trachea was clearly divided into membranous and cartilaginous portion. The principal bronchi were lined each with a layer of columnar ciliated epithelial cells and below the epithelium was a layer of smooth muscle. The respiratory bronchioles were lined by a low columnar epithelium and continued as alveolar ducts and opened into alveolar sacs. The average diameter of trachea, respiratory bronchiole, alveoli duct and alveoli were $2.82 \pm 0.09 \mu$ m, $0.50 \pm 0.04 \mu$ m, $0.47 \pm 0.02 \mu$ m and $0.17 \pm 0.01 \mu$ m respectively. Our findings were compared with similar report in other mammals. This study will help to bridge the dearth of information in this area.

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1. Introduction

Bats are unique among mammals in their capacity for continuous flapping flight. Wind tunnel experiments have shown that flapping flight is energetically a very demanding form of exercise, being substantially beyond the energetic capacity of terrestrial mammals of similar size during maximum exercise (Maina, 2000a).

Bats are the second largest Order (Chiroptera) of mammals in the world and the only mammal capable of true flight (Bat Conservation International, 1997). They are grouped into two Suborders: Megachiroptera (megabats) and Microchiroptera (microbats). The straw-colored fruit bat (*Eidolon helvum*) is the second largest fruit bat in numbers on the African continent. The bat weighs approximately 250–310 grams and has an average wingspan of 80 cm (Bergmans, 1990).

E. helvum has a strong flyer built for endurance rather than mobility, and its body structure supports long migrations (Thomas, 1983). They are unique in their vagility (potential for long-distance travel) and this activity may aid in spread of diseases (Turmelle and Olival, 2010). Bats have been shown to increase their oxygen consumption during sustained flight by at least 20 times compared to other mammals (Maina, 2000a). In general, the energetics of bat flight appears to be comparable to that of birds (Jurgens et al., 1981).

In contrast, birds have evolved voluminous air sacs which continuously and unidirectionally ventilate the compact, rather rigid lung (Maina, 2000b). In active animals, the increased metabolic rate during locomotion places great demands on the respiratory system (Bundle et al., 1999). Flapping flight (active - powered) is energetically a very costly form of locomotion (Maina, 2000a). Animals with high energetic requirements have physiological and structural compromise solutions for the required energetic efficiency, environmental demands and lifestyle constraints (Schmidt-Nielsen, 1997). Because flight is one of the most energetically expensive forms of locomotion, these compromises are particularly evident in flying animals.

The general anatomy of the respiratory tracts of bats is, of course, fundamentally similar to that of terrestrial mammals (Maina, 2000b).

Bat lungs are similar to those of non-flying mammals, but may be highly refined to operate at near maximum values and this has rekindled research interest in the area in search of microscopic differences between the lung of bats and other mammals.

The knowledge of anatomy is paramount for better understanding of the straw coloured fruit bat. Despite having an essentially mammalian type of lung, bats nevertheless appear to have an exercise capacity as good as that of birds. So there is every possibility that bat lung might possess structural adaptations that could help in the above functions. Several investigators have made pioneering efforts to characterize various systems of this bat in Nigeria; female reproductive system (Samson et al., 2009) and digestive system (Ofusori et al., 2008) of this bat.

Further, the histology of the lower respiratory tract in other mammals such as the laboratory rat (Choi et al., 2000; Widdicombe et al., 2001), hamster (Kennedy et al., 1978), squirrel (Nakakuki, 1979) and guinea pig (Kliment et al., 1972) have been described in detail. However, such description in the straw coloured fruit bat is lacking. Various studies have been performed on gross morphology, morphometry and comparative respiratory morphology (Maina, 2000b) no work has been conducted on the microscopic anatomy of the trachea, bronchi and lungs of lower respiratory tract of the bat. Therefore, the present study was aimed at documenting the microscopic features of the trachea, bronchi and lungs of the straw coloured fruit bat found in Nigeria. This study will serve as a fulcrum for future biomedical investigations regarding the respiratory system of the straw coloured fruit bat and it will help to bridge the dearth of information in this area.

2. Materials and methods

2.1. Study design

Seven straw colored fruit bats with an average weight of 250g were utilized in this study. The bats were captured live during the daytime from Zuru Local Government Area in the Southeastern part of Kebbi State located in Northwestern Nigeria. Zuru lies between the latitudes 11°15' to 11°56'N and longitudes 4°03' to 5°25'E. It falls within the Sudan savanna type of vegetation with grasses giving up to 1-1.5m in height and feathery. The mean annual rainfall is about 600mm to 1000mm and the mean minimum and maximum temperatures were 27°C and 39°C respectively (Yatswako et al., 2007).

The capturing process was done during daytime at their roosts using hand nets with extendable poles. The bats were held in plastic cages (2.9 m long × 2.4 m wide × 2.0 m in height). The flight cages were exposed to natural conditions and several leaves were hung on the ceilings of the cage to provide roosts for the bats. After the capture process, the bats were fed with bananas, mango and water ad libitum.

The bats were transported by road to the Department of Veterinary Anatomy laboratory, Faculty of Veterinary Medicine, Usmanu Danfodiyo University, Sokoto, Nigeria

2.2. Bat identification

The bats were identified at the Zoological laboratory unit, Department of Biological Science, Faculty of Sciences, Usmanu Danfodiyo University, Sokoto, Nigeria.

2.3 Sample collection

The bats were euthanized severing the jugular vein. The ventral cervical region and entire thoracic cavity were exposed by dissection. The lower respiratory tract was exteriorized and freed from other thoracic viscera. The trachea, principal bronchi and lungs were separated. The tissue from trachea and principal bronchi were used sectioned for histomorphology. Lung tissues from the same seven samples were utilized to study the normal histological features of the lungs. Sections were stained with the conventional Haematoxylin and Eosin (H & E) stain.

2.4. Tissue processing

The tissues were fixed in 10% phosphate-buffered formalin for three days. Thereafter, the samples were washed in water and kept therein for twenty four hours. The tissues were processed to obtain 5µm thick paraffin sections as described earlier (Maina, 2002) by using Rotary microtome (Model 42339, Berlin, Germany). The Paraffin sections were stained with Haematoxylin and eosin (H and E).

2.5. Histomorphometric analysis

Histological measurements were recorded on the lower respiratory system portion of the bats. This included the average diameter of tracheas (n=7), respiratory bronchioles (n=14), alveoli ducts (n=20) and alveoli sacs(n=40). The diameters of the above structures sections were obtained with the light binocular microscope (Lieca®, Germany) with the aid of a digital eyepiece (Motic cam® DCM100, Resolution: 1.3M pixels, England). The respiratory bronchioles and alveoli ducts were defined by the fact that the former does not give off alveolar sacs unlike the latter. Photographs were taken using a digital camera (7.5mega pixels).

3. Results

The trachea of the bat was clearly divided into the membranous and cartilaginous portions, with its mucosa lined by a respiratory epithelium composed of pseudostratified ciliated columnar cells on a basal lamina followed by the trachealis muscles closely attached to the cartilageous ring (Fig. 1 and 2).

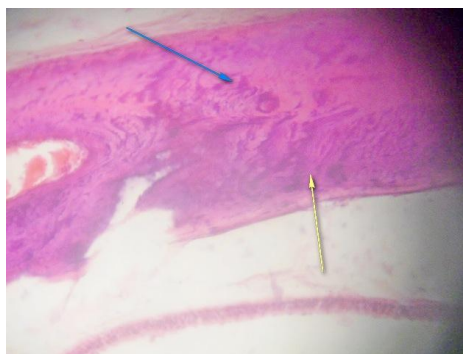


Fig. 1. Photomicrograph of the region transverse section (yellow arrow cartilaginous Muscle Blue arrow cartilage) (H. & E. x100).

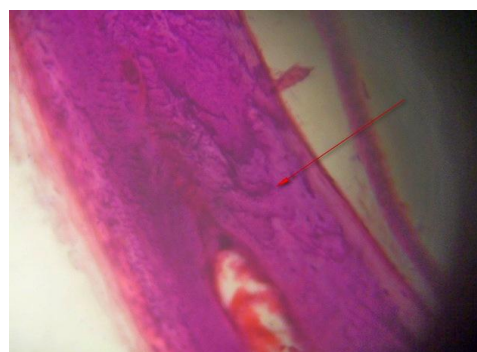


Fig. 2. Photomicrograph of the of the tracheal membranous region of the tracheal trachealis transverse section red arrow (H. & E. x100).

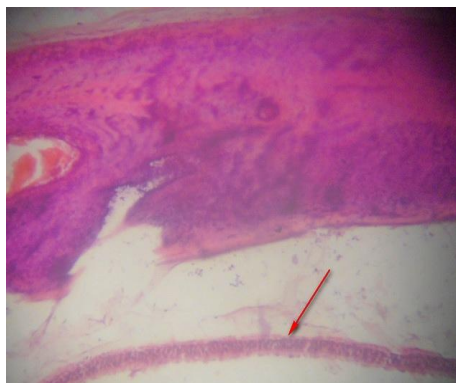


Fig. 3. Photomicrograph showing the tracheal epithelial lining type (Red arrow for the epithelia lining) transverse section (H. & E. x400)

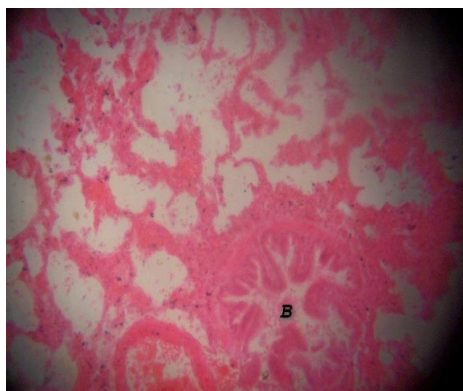


Fig. 4. Photomicrograph of the respiratory labeled B (H. & E. x100).

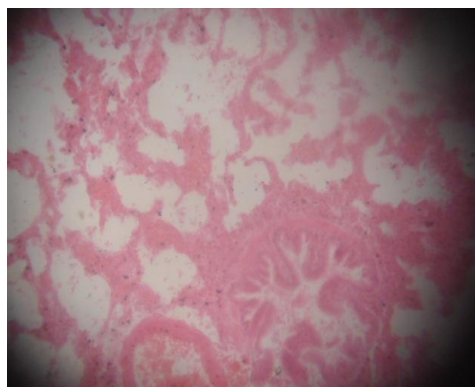


Fig. 5. Photomicrograph of the bronchi labeled B (H. & E. x100).

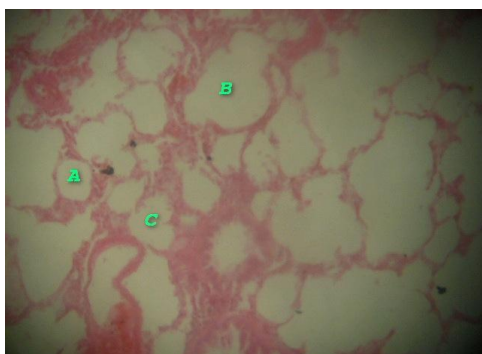


Fig. 6. Photomicrograph of the lung tissue of the Straw coloured fruit bat, illustrating the some structures of the transitional component.

A: Alveoli sacs.

B: Respiratory bronchiole.

C: Alveoli duct. (H. & E. x100).

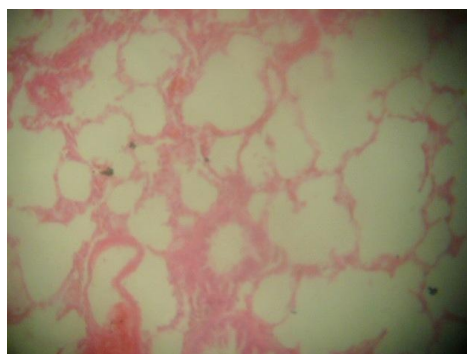


Fig. 7. Photomicrograph of the lung tissue of the Straw coloured fruit bat showing the alveola sacs (H. & E. x100).

In the present investigation, the left and right principal bronchi were lined with a layer of columnar ciliated epithelial cells and below the epithelium was a layer of smooth muscle. Several short lobar bronchi branched off from the main bronchi. They consisted of cuboidal epithelium and a layer of one or two smooth muscles and opened into segmental bronchi and respiratory bronchioles (Fig. 3). The respiratory bronchioles were lined by a low columnar epithelium with thin lamina propria and continued as alveolar ducts and the alveolar ducts opened into numerous alveolar sacs (Fig. 4-7).

3.1. Histomorphometric assessment

The result of the average diameters of the tracheal, respiratory bronchioles, alveoli duct and alveoli sac were represented in Table 1 with the number of counted structures in parentheses

Table 1

Sizes of airway structures in the straw coloured fruit bat.

Structure	Mean \pm SEM(μm)	Min	Max
Tracheal diameter (n=7)	2.82 \pm 0.09	2.5	3.1
Respiratory bronchioles diameter (n=14)	0.54 \pm 0.04	0.4	1
Alveolar duct diameter (n = 20)	0.47 \pm 0.02	0.3	0.6
Alveoli sac diameter (n = 40)	0.17 \pm 0.01	0.1	0.3

4. Discussion

The histological arrangement of the lower respiratory tract in bats was similar to other mammals despite ability to fly and the absence of accessory structure like the air sac in bats (Bank, 1993). The structure of the lung is adapted to meet the oxygen demands of an animal, which in turn will reflect to various factors such as body weight, wing web and mode of life. Bats are known to be excellent fliers thus; much demand is being placed on the lung to supply the much needed oxygen (Maina, 2000a).

The mucociliary nature of the tracheal and bronchial epithelium is very important in respiration in bat since one of the major functions of tracheal and bronchial epithelium is clearance of inhaled harmful particles (Banks, 1993). According to Banks (1993) this important lung-protective function depends on the epithelial lining types, epithelial types were similar to those observed in this study.

In this study, the average diameter of the trachea was 2.82 \pm 0.09 μm , which was lower when compared to the finding of Widdicombe et al., (2001) in other mammal like the hamster, rat and guinea pig with an average tracheal diameter of 1.5 μm , 2.5 μm and 2.5 μm respectively. Choi et al., (2000) recorded a positive relationship between tracheal diameter, bronchi diameter and number of mucous glands in mammals and concluded that increased size of conductive airway (by increased tracheal diameter and bronchi diameter) resulted in an increased inflow velocity of inspired air which culminated in an increased rate of foreign body deposition.

The tracheal epithelia recorded in the present study were similar to that of the rodents as reported by Reznik (2004). However, Kennedy et al. (1978) stated that the respiratory epithelia in the Syrian golden hamster ranged from ciliated pseudostratified columnar to low columnar epithelium with goblet cells. This is also similar to the result of the present study in which respiratory epithelium in sagittal sections of the trachea revealed similar pattern of epithelia lining.

Our finding is similar to what is obtainable in guinea pig as reported by Dalen (1983) in respect to the incomplete hyaline cartilage ring as observed in this study.

The average value of the alveoli diameter seen in this present study is 0.17 \pm 0.01 μm which is contradictory to the findings of Kennedy et al., (1978) where they recorded an average alveoli diameter of 6 μm in the adult golden Syrian hamster. They also recorded an average diameter of 13 μm and 16 μm for the alveolar duct and respiratory bronchioles respectively which was also not in agreement with our findings. The values reported by Kennedy et al., (1978) are higher, compared to the values we obtained in bats which could be attributed to the body size of the bat.

In this studies, parabronchi were absent in the bats which is in contrast with what is obtainable in the lung of birds, though both of them are flying animals but with little variation with respect to some anatomical structures.

5. Conclusion

In the present study, histological features of the lower respiratory system in the Straw coloured fruit bat were observed. It was suggested that the lower respiratory system in the fruit bat was almost similar to that of the hamster and some other mammals except for the smaller sizes of the alveoli, alveolar ducts and respiratory bronchi, when compared to those of the Syrian golden hamster. Thus, the lower respiratory system in the Straw coloured fruit bat can be used as a model of the rodent respiratory system histology in phylogenetic studies.

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