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Spermatogenesis in Schlieffen's bat, *Nycticeius schlieffenii* (Chiroptera: Vespertilionidae)

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Abstract

Investigated spermatogenesis in Schlieffen's bat, *Nycticeius schlieffenii*. Schlieffen's bat exhibits an extended spermatogenesis with the various cell stages easily identifiable in the sequence of events. From late September to October there is only a slow increase in type A spermatogonia while proliferating type B5 spermatogonia predominate in the seminiferous tubules during the period November–February. December–February can be regarded as the time when change over to primary spermatocytes occurs, although at a very slow rate. March is an active month when all spermatogenic cells can be found. Spermatozoa only start to accumulate in the epididymes in April (mid-autumn), i.e. about three months before the first copulations.

Introduction

Schlieffen's bat is one of the smaller South African bat species, with males averaging 4.7 g and females 5.1 g (Rautenbach 1982). It has a distinct easterly distribution within South Africa, but is one of the more common species of bat within its range. *N. schlieffenii* does not migrate seasonally, as they were collected from the same area throughout the year. During the day this species roosts at sites such as hollows in trees (Verschuren 1957) or rock crevices (Pienaar et al. 1987).

Copulation commences during June and the females have spermatozoa in the uterine horns from then until the end of August when ovulation occurs. These bats are seasonally monoestrous with births occurring during November (van der Merwe and Rautenbach 1987).

The purpose of this study is to describe the male reproductive cycle.

Material and methods

Specimens of Schlieffen's bat were collected at monthly intervals with macro-mistnets (Rautenbach 1985) at several localities at Pafuri (22° 25′ S; 31° 12′ E) in the northern region of the Kruger National Park, Eastern Transvaal lowveld. At least five male specimens were collected for each calender month. At the field laboratory the bats were sacrificed with technical ether. The testes were dissected out and preserved in Bouin's fixative for histological examination. Following routine paraffin wax embedding, all testes and epididymes were sectioned at 5 μm, mounted and stained with Ehrlich's haematoxylin and eosin.

Results

During August (end of winter) the testes were inactive with no sign of spermatogenic activity in the seminiferous tubules although some spermatozoa from the previous cycle could still be found in the caudae epididymides. The relatively large lumina of the

seminiferous tubules were lined with a compact layer of Sertoli cells and only occasional stem spermatogonia could be found against the basal lamina (fig. 1). At this stage the Leydig cells were small and compact.

The first signs of spermatogenic activity were towards the end of September when a few spermatogonia undergoing mitosis were found. Within the epididymes only cell debris could be found with no sign of spermatozoa. Leydig cells were similar in appearance to the previous month.

Towards the end of October (mid-spring) the number of type A spermatogonia had increased considerably resulting in the formation of a near complete layer of cells on the inside of the basal lamina (fig. 2). A few type B spermatogonia were also present. The Leydig cells were still compact.

By the end of November a marked increase in type B spermatogonia was found (fig. 3) while the Leydig cells were still compact.

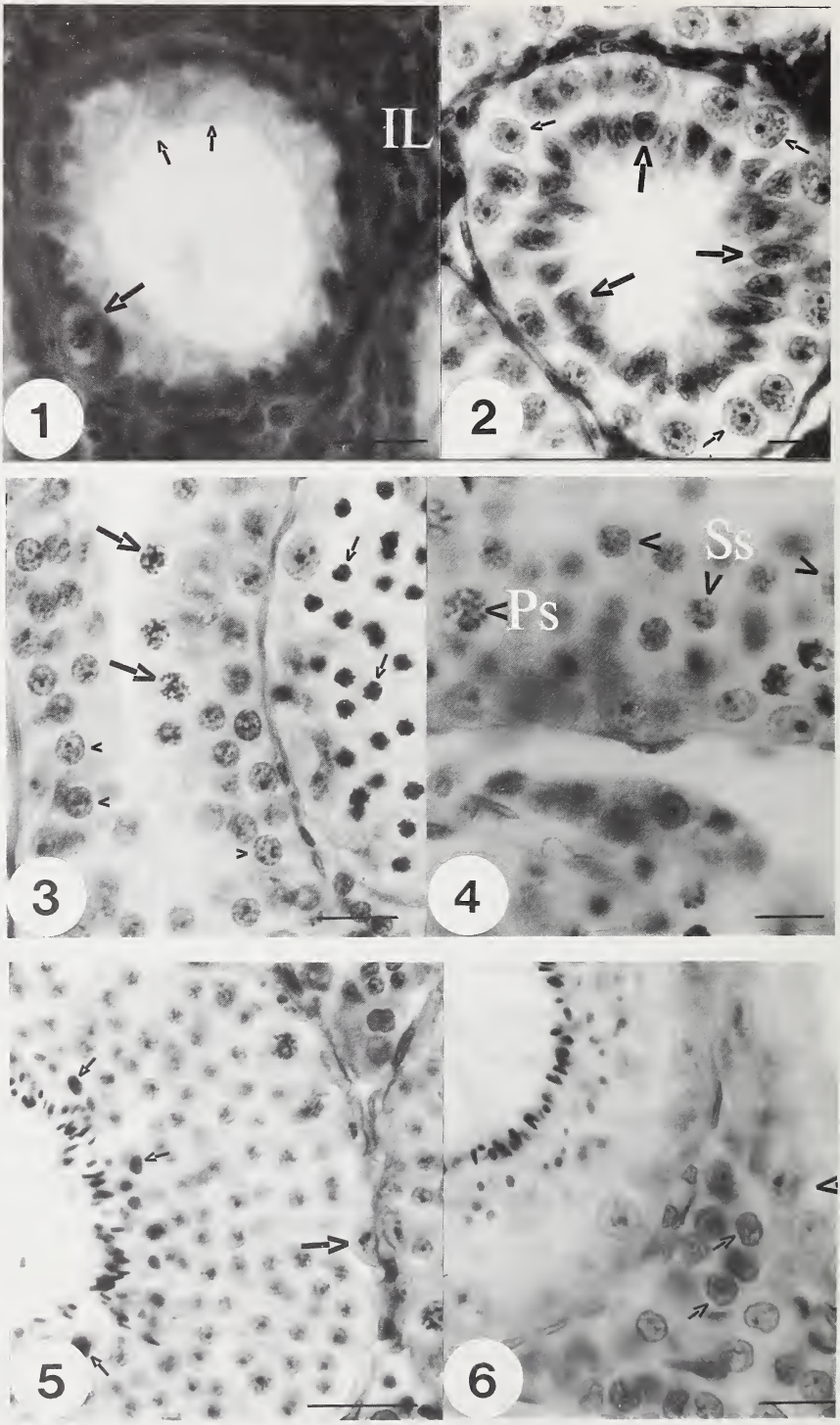
Towards the end of December resting type A spermatogonia, many type B spermatogonia (both undergoing mitosis) and a few primary spermatocytes could be found (fig. 3). The Leydig cells were still compact although some showed vacuoles.

From January towards the end of February the Leydig cells were swollen with vacuoles. During both months many type B spermatogonia were present, the majority had completed the mitotic divisions, and were in the growth phase. Primary spermatocytes were still scarce at the end of January, but they were more numerous towards the end of February.

From the end of February to the end of March nearly all stages in germ cell development could be found including the short lived secondary spermatocytes (fig. 4). During this period spermatozoa were still absent from the epididymes. Primary spermatocytes and spermatids were abundant with large numbers of type B spermatogonia in the growth phase. The first time that spermatids with condensed nuclei (undergoing spermiogenesis) and a few spermatozoa were found, was at the end of March and beginning of April. At this stage, however, they were still very scarce.

Towards the end of April primary spermatocytes and spermatids with condensed nuclei were abundant. Numerous spermatozoa were attached to Sertoli cells or accumulated in the lumina of the seminiferous tubules while large numbers were also present in the epididymes. There was a drastic decrease in the number of type B spermatogonia while against the basal lamina only a few resting type A spermatogonia could be found (fig. 5). At this stage spermiogenesis was actively taking place with cytoplasmic residual bodies very conspicuous (fig. 5). The caudae epididymides were crowded with spermatozoa while the Leydig cells were swollen.

Between May and June (beginning of winter) the situation changed with a decrease in the number of primary spermatocytes. Spermatids and spermatozoa were abundant and the caudae epididymides crowded with spermatozoa. A few individual type B spermatogonia were still present. Spermiogenesis was continuing with an abundance of cytoplasmic residual bodies. Although the majority of Leydig cells were still swollen, some of them were becoming compact with darker, more unevenly shaped nuclei. At the end of July the seminiferous tubules were exhausted. With the exception of some spermatozoa, and a few resting (stem) type A spermatogonia, no spermatogenic cells could be found (fig. 6). In the majority of seminiferous tubules the nuclei of the Sertoli cells were lying against the basal lamina with no spermatogenic cells present except individual stem type A spermatogonia. The caudae epididymides were still crowded with spermatozoa. Most of the Leydig cells were compact with shrunken nuclei of uneven dimensions.



Discussion

Vespertilionid bats in the temperate zones generally copulate during autumn with the spermatozoa being stored in the female uterus until the following spring when ovulation and fertilization occur (Racey and Potts 1970). In the subtropical climate of Pafuri, copulation in Schlieffen's bat is initiated towards the end of June (early winter) with the first ovulations towards the end of winter (Van der Merwe and Rautenbach 1987). Spermatozoa were found in the uterus of females collected at the end of June and end of July while four out of five females collected at the end of August were pregnant (Van der Merwe and Rautenbach 1987). At present it is not certain whether the females store spermatozoa during June till August, as males have epididymes crowded with spermatozoa during the same period. At present it appears likely that frequent copulations occur during this period.

From the same study area van der Merwe and Rautenbach (1989) have also found prolonged spermatogenesis in the lesser yellow house bat *Scotophilus borbonicus*. The present study agrees with their findings that spermatocytogenesis is extended but not spermiogenesis.

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Zusammenfassung

Spermatogenese bei Schlieffens Fledermaus, Nycticeius schlieffenii (Chiroptera: Vespertilionidae)

Untersucht wurde die Spermatogenese bei Schlieffens Fledermaus *Nycticeius schlieffenii*. Vom späten September bis Oktober vermehren sich Typ-A-Spermatogenien nur langsam, während von November bis Februar Typ-B-Spermatogenien, die sich rasch vermehren, in den Hodentubuli vorherrschen. Von Dezember bis Februar beginnt – allerdings in geringem Umfang – die Bildung primärer Spermatozyten. Im Monat März sind alle spermatogenen Zellen vorhanden, und im April (in der Mitte des Südherbstes), d. h. etwa drei Monate vor den ersten Paarungen, beginnen sich Spermatozoen im Nebenhoden anzuhäufen.

Fig. 1–6. Sections through parts of testes collected during different months. *Fig. 1.* At the end of August showing a seminiferous tubule lined with Sertoli cells (small arrows) and one stem spermatogonium (large arrow). The interstitial cells of Leydig (IL) can be seen between the tubules. Bar = 25 μm. – *Fig. 2.* At the end of October showing a seminiferous tubule with type A spermatogonia (small arrows) against the basal lamina and the nuclei of the Sertoli cells (large arrows) on their inside. Bar = 25 μm. – *Fig. 3.* At the end of December showing numerous type B spermatogonia undergoing mitosis (small arrows), with mature type B spermatogonia (arrow heads) and a few primary spermatocytes (large arrows). Bar = 25 μm. – *Fig. 4.* At the end of March showing primary spermatocytes (Ps) and secondary spermatocytes (Ss). Bar = 10 μm. – *Fig. 5.* At the end of April showing a seminiferous tubule with masses of spermatozoa and of cytoplasmic residual bodies (small arrows). Against the basal lamina a few resting type A spermatogonia can be seen (large arrow). Bar = 25 μm. – *Fig. 6.* At the end of July showing one seminiferous tubule with numerous spermatozoa and cytoplasmic residual bodies while the adjacent one is empty with only Sertoli cells (large arrow) lining the basement lamina. The uneven shape of the nuclei of the interstitial cells of Leydig is indicated with small arrows. Bar = 15 μm

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