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Host-parasite Relationships of Acarine Parasites and their Vertebrate Hosts.

I. The Lesions Produced by Bakerocoptes cynopteris in the Skin of Cynopterus brachyotis

By M. M. J. LAVOIPIERRE, C. RAJAMANICKAM and P. WARD

Introduction

This paper is the first of a series which will be devoted to a study of the relationships between "host-dwelling" acarine parasites and their vertebrate hosts. A "host-dwelling" parasite (see Audy, 1948, 1958 1), may be defined as a parasite living permanently on its host where it feeds, reproduces and finds shelter.

Many species of host-dwelling mites have been described from a wide variety of vertebrate hosts, including amphibians, reptiles, birds and mammals. They range from superficial parasites which tunnel in the integument, such as the ophioptids of reptiles and the sarcoptids of mammals, to species which invade the respiratory tract, examples of which are the cytoditids of birds and the pneumonyssids of mammals.

In this and subsequent papers attention will be focussed on acarines which live on or in the integument of their hosts. While emphasis will be placed on the nature of the reaction of host tissues to invasion by mites, some consideration will be given to the parasite. Whenever a new species of mite is mentioned, a brief description will be given, with the formal description to follow elsewhere.

The present contribution is devoted to a study of the cyst-like swellings produced by a sarcoptiform parasite in the wing membrane of the fruit bat *Cynopterus brachyotis*. The parasite was described by Fain in 1962 from material taken from a bat collected on Pagi Island, Malaya, in 1902 and preserved in the United States National Museum. The mite was named *Bakerocoptes cynopteris* and ascribed to the family Teinocoptidae.

When we discovered the parasite, which is relatively common on *C. brachyotis*, we were not aware of Fain's description. Our first impression was that it was a teinocoptid. However, it showed so many distinct characteristics, particularly in its ecological relationships, that we felt it should be classified in a group close to, but distinct from, the family Teinocoptidae. We have since noted that Fain was equally puzzled by its true relationships, creating a separate subfamily to accommodate the mite. We feel that since *B. cynopteris* is so peculiar in many respects, it should be placed in a distinct family, the Bakerocoptidae.

¹ AUDY has grouped parasites as (1) food-dwelling, (2) host-dwelling, (3) nest-dwelling and (4) field-dwelling.

Review of the Literature

Little is known of the histopathological reaction of the host's skin to any of the many species of parasitic mites described from wild mammals and birds. While it is true that many parasitic acarines do not appear to elicit any response, in others the reactions are so striking as to be plainly visible (Lukoschus, 1962; Ward, Lavoipierre & Rajamanickam, 1965). Most papers on skin mites have been concerned only with species descriptions. Notes on tissue reactions are generally limited at the most to a brief paragraph, often to a mere sentence. Even in the case of animals showing gross clinical changes, many interesting lesions have been summarily described without reference to histological changes.

Within recent years, however, there have appeared a few papers which undoubtedly merit a special place in the literature of the host-parasite relationships of wild animals and their mite parasites. Clark & Stotts (1960) have published an interesting note on the lesions caused by the trombiculid Whartonia in the skin of wild ducks. Daniel & Slais' (1957) report of the skin-penetrating habit of Euschongastia ulcerofaciens in wild rodents, illustrated with excellent photographs, is a most useful contribution. Hyland's (1961) fine experimental study of the development of the larva of the chigger Hannemania hegeneri in the skin of amphibians is a model for future workers. More recent works on chiggers are the papers of Hoeppli & Schumacher (1962) and Schumacher & Hoeppli (1963).

Amongst the sarcoptiforms, studies of host-parasite relationships—with particular reference to tissue changes—have been more limited. Those of Grunberg & Kutzer (1962a, b) and Kutzer & Grunberg (1963) on feather mites of birds are useful. Böhm & Supperer's (1958) study of the listrophorid *Myocoptes tenax* on *Microtus agrestis* gives much useful histopathological data.

Little is known of the reaction of the skin of bat infestations with acarines, though early studies of Rodhain (1923, 1926) give a good account of the lesions caused by the sarcoptid *Nycteridocoptes pteropi* Rodh. & Ged. on *Eidelon helvum* Kerr. Unfortunately, Rodhain supplies no detailed histological picture. A brief description is given of the lesions provoked by *Teinocoptes epomophori* in the wing membrane of *Epomophorus wahlbergi haldemani* and figures are provided showing the epidermal cupules produced by the female mites. Other studies are those of Roessler (1932) and Vitzthum (1932) on the lesions produced by

Notoedres roessleri in the bat Myotis nigricans and that of VITZ-THUM (1943) on mange caused by Nycteridocoptes poppei. Lukoschus (1962) gives some brief illustrated notes of the changes taking place in the wing membrane of European bats infested with N. poppei. Rack (1962) has provided some excellent photographs of cysts produced by N. poppei in the wing membrane of Myotis myotis.

Materials and Methods

The species of bat used in the study was the Dog-faced Fruit-bat, Cynopterus brachyotis (order Chiroptera, sub-order Megachiroptera), one of the commonest mammals in Singapore (Fig. 1). During the day these small (80–90 mm body length) fruit-bats hang in clusters under the eaves of houses, or some other sheltered position. They emerge at dusk to spend the night feeding on the ripe fruit of a variety of shrubs and trees.



Fig. 1. C. brachyotis (the Dog-faced Fruit-bat), the host of B. cynopteris.

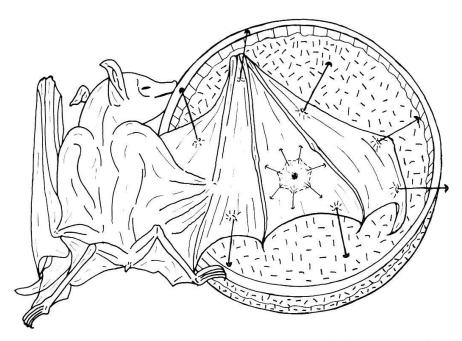


Fig. 2. Showing the method of preparing a cyst of Bakerocoptes for fixation. The wing is spread out over a petri dish containing a piece of cork. The tissue immediately surrounding the cyst is pinned out, and then cut out from the membrane, along the dotted line.

The bats bearing the mite-containing cysts were captured in a suburban garden in Bukit Timah, Singapore, from October 1964 to November 1965. A single mist-net, measuring 10×2 metres, erected at ground level was used to catch the bats, as many as twenty bats being netted in a night. Though mist-nets are designed primarily for bird trapping, they work admirably with fruit-bats, which navigate by sight and not by echo-location. The bats were not damaged in any way by the nets and were usually left till the following morning before being removed. Some of the animals were killed with chloroform immediately after removal from the nets, and taken to the laboratory for examination. Others were kept alive for subsequent investigation.

The cysts on infected bats were studied in two ways: Some of the cysts and the immediate surrounding tissue were cut out of the wing membrane and introduced individually into hollow-ground watchglasses containing lactochlorophenol (GORDON & LAVOIPIERRE, 1962). When the tissues had softened, the cysts were carefully dissected open and the enclosed mites, comprising adults and immature stages, released in the lactochlorophenol. A number of the mites were permanently mounted in Ewing's modification of Berlese's gum chloral mountant.

Secondly, tissues from some of the infested bats were fixed in Bouin's alcoholic fixative. We found that in order to obtain good sections, the best way of doing this was as follows (see Fig. 2). A thin, flat piece of cork was firmly wedged in a petri-dish and the wing membrane of a recently killed bat bearing the cysts spread over the dish. The membrane surrounding the cyst was then carefully pinned out on the cork in such a way so as to gently stretch a roughly circular piece of skin with the cyst at its centre (Fig. 2). By cutting outside the ring of pins, the circle of tissue was left attached to the cork in the dish which could then be flooded with fixative and covered to prevent evaporation. All such tissues were fixed for about 18 hours and subsequently sectioned and stained with haematoxylin and eosin.

Results

a) Distribution of cysts in bats

We have examined *Cynopterus brachyotis* taken in Singapore and in the Mersing district of Johore (Malaya) and have found affected animals from both localities. Other species of bats captured have never had any cysts on the wing membranes. These include several hundred specimens of *Scotophilus temmincki* from Johore and Singapore; many *Cheiromeles torquatus* from Singapore and Malaya and *Hipposideros* spp., from Malaya; and a considerable number of other, unidentified, bats.

TABLE 1

Showing the numbers of *Cynopterus brachyotis* examined during an 8-month period and the numbers bearing cysts of *Bakeroptes cynopteris*. Cysts have been found only in the wing membranes, never on the hairy parts of the animal

Batch No.	Date of capture	No. of bats examined	No. of bats infested	No. of cysts
1	24. 3. 1965	10	2	2 (each bat with 1 cyst)
2	21. 6. 1965	15	1	3
3	30. 6. 1965	15 (3 were juveniles)	1	6
4	5. 8. 1965	5	0	_
5	19. 8. 1965	12	1	1
6	4. 11. 1965	3	1	5
7	22. 11. 1965	15	1	1

Very young individuals of *C. brachyotis* do not appear to bear cysts, these having only been found in young and mature adults, both male and female. Details were not kept of all the bats examined, but in Table 1 is given the incidence of infestation in 7 batches of bats, a total of 75 animals. Of the 75 bats, 7 (9%) bore cysts. The largest number of 'ripe cysts' found on an in-

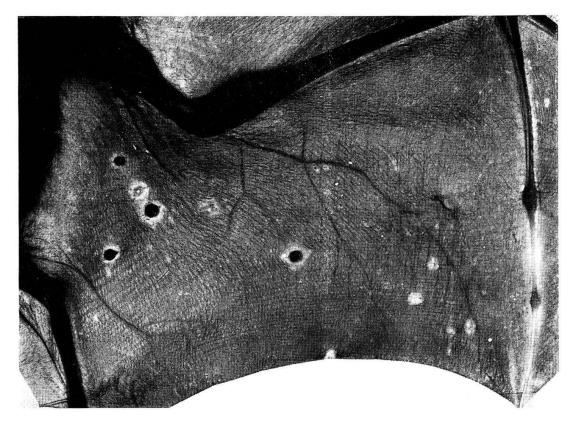


Fig. 3. Transilluminated wing of C. brachyotis showing active and burnt out cysts of B. cynopteris.

dividual bat was 7 (not recorded in Table 1); five were on the right wing and 2 on the left wing. There is some evidence that a bat infested with *Bakerocoptes cynopteris* may bear many more than 7 cysts (see Fig. 3). In Figure 3, the transilluminated right wing of an infected bat is shown. The dark spots surrounded by a halo are occupied cysts whereas the light areas reveal the position of former cysts. In at least two areas scarring is still evident but towards the periphery of the wing the scarred tissue has disappeared leaving translucent spots.

b) Lesions caused by Bakerocoptes cynopteris in the wing of its host

Gross appearance of the lesions. If the wing of a bat infested with *B. cynopteris* is extended, the cysts, although relatively small in size (fully developed cysts measured 1.5 to 2 mm in diameter), attract attention because of their distinctive appearance (Fig. 4). Each cyst appears as a discrete, circular, pea-like, nodular swelling, the dark colour of which contrasts sharply with a circumvallating zone of pale tissue translucent to transmitted light (Fig. 5). The lighter zone in its turn merges imperceptibly with

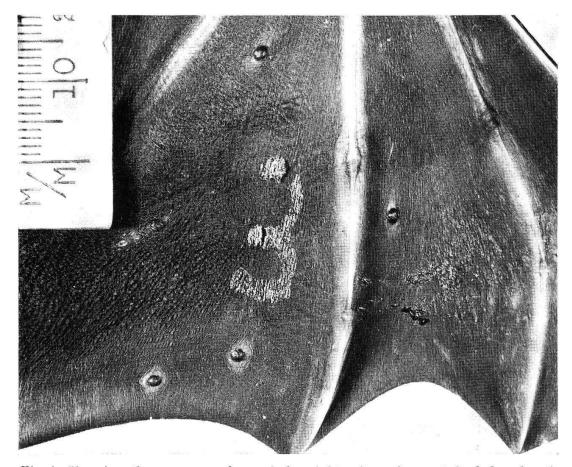


Fig. 4. Showing the upper surface of the right wing of an adult C. brachyotis with five cysts of B. cynopteris.

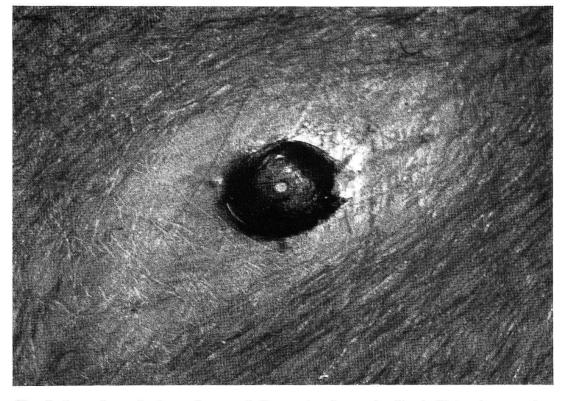


Fig. 5. An enlarged view of one of the cysts shown in fig. 4. Note the opening in the centre of the cyst.

the neighbouring wing tissue giving the cyst the appearance of being surrounded by a halo. Every cyst is provided with a single, centrally placed, punctiform orifice in most cases on the upper surface of the wing. Through the circular opening, with its slightly raised rim, the anal end of the female mite can readily be seen. The side of the cyst bearing the opening is distinctly more raised, and more conspicuous, than that of the opposite surface.

Histological appearance of the cyst (Figures 6, 7, 8 and 9). Sections cut through the middle of the cyst, show a striking resemblance to retention or implantation cysts, except for the contained parasites. The cyst lies entirely within the tissues of the wing membrane, enclosed by the upper and lower epidermis, which is

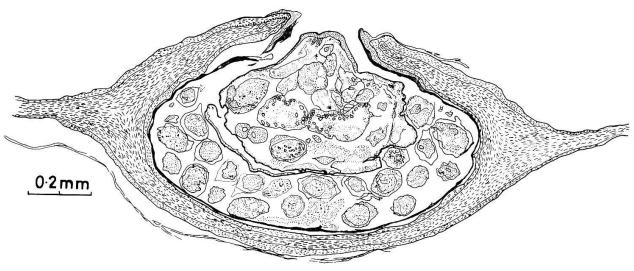


Fig. 6. A drawing of a histological section cut through a "ripe" cyst of B. cynopteris. See fig. 7 for an explanation of the various structures shown.

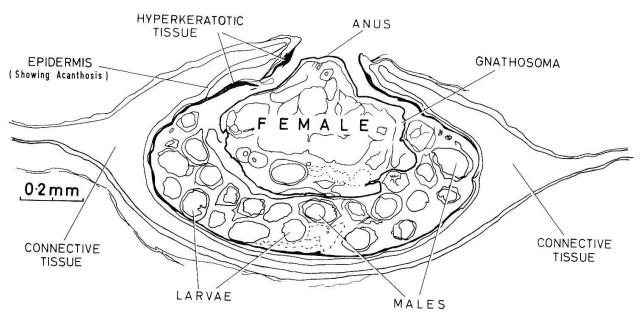


Fig. 7. A tracing of fig. 6 with a key to the structures illustrated.

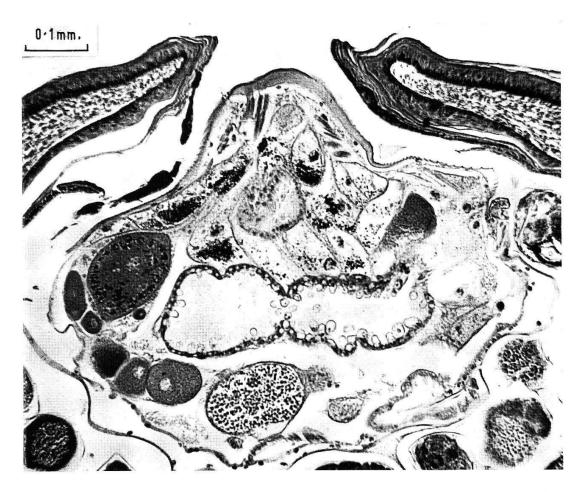


Fig. 8. A photograph of the upper portion of a section through a cyst, showing the cyst orifice and a female B. cynopteris lying below it.

considerably thickened at the point where the cyst is located. The cyst thus consists of a capacious, epithelium-lined chamber, the cavity of which leads to the exterior through a small, centrally-placed opening. The epithelium lining the cyst is continuous, through the opening, with that of wing membrane. The connective tissue in which the cyst is embedded shows a marked hyperplasia.

Changes in the epithelium. In the normal condition the epidermis of the dorsal and ventral surfaces of the wing is thin (Fig. 10), the two epidermal layers being separated by a lamella of connective tissue which may be regarded as a conjoint dermis.

Each epidermal layer appears to be composed of a single layer of somewhat flattened cells supporting a narrow zone of stratum lucidum on which rests a very thin stratum corneum. The dorsal epidermis is heavily charged with melanocytes and is opaque in appearance. The much lighter colour of the ventral epidermis is due to a relative scarcity of melanocytes which are sparsely distributed amongst the non-pigment-producing cells.

As the normal epithelium of each surface begins to merge with

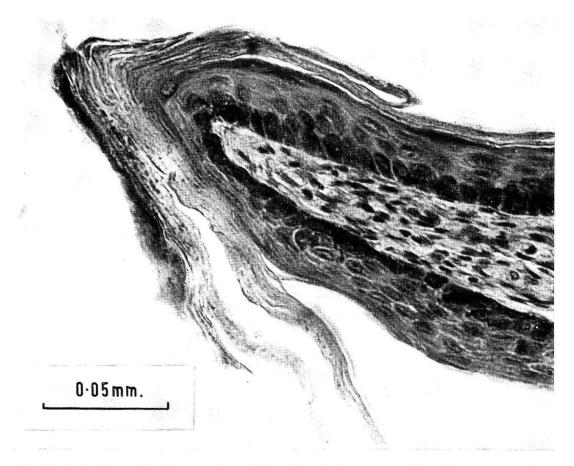


Fig. 9. A photograph of a section of the rim of the cyst-orifice. Note the marked acanthosis and hyperkeratosis. This should be compared with fig. 10 which is photographed at almost the same magnification.

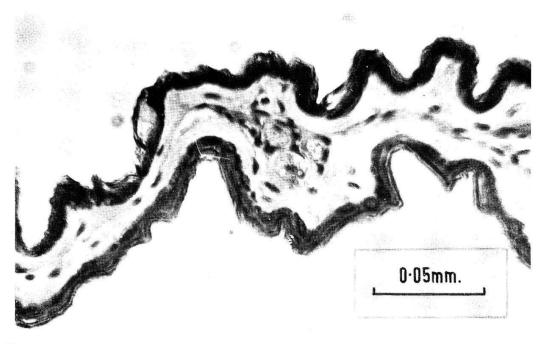


Fig. 10. Photograph of a section through the normal wing membrane of B. cynopteris. Note the upper epidermis is more pigmented than the lower epidermis. The epidermis becomes greatly thickened in the region of the cyst (see fig. 9).

that covering the cyst it shows a progressive thickening due to an increase in the number of cells, and becomes differentiated into several clearly recognisable strata (Fig. 9). The most striking change in the epithelial layers over the cyst is a moderate but distinct acanthosis, unaccompanied by hyperkeratosis. In the immediate neighbourhood of the cyst opening, however, there is a definite hyperkeratotic reaction which becomes pronounced within the neck of the cyst (Fig. 9). This reaction is continued, though to a diminished extent, over the acanthotic epithelium lining the cyst wall.

At its point of maximum development, in the neighbourhood of the cyst mouth, the epithelium of the dorsal surface consists of four to five layers of cells. The basal layer of columnar cells (the stratum germinativum) contains many melanocytes. This is followed by a well formed malpighian zone, composed of 3-4 layers, throughout which are distributed aggregations of melanotic granules. The thin stratum granulosum and stratum lucidum are overlaid by a slightly thicker stratum corneum.

Within the neck of the cyst there is a progressive flattening of all the epithelial cells, a process which becomes fully developed within the wall of the cyst. The melanotic basal layer loses its columnar appearance. Hyperkeratosis is well marked, and some of the mites contained in the cyst appear to burrow in the thickened stratum corneum, giving this tissue the appearance of being loculated.

The epithelium of the ventral wing surface beneath the cyst also shows well-marked changes similar to those of the dorsal surface. The basal layer, however, is provided with only a few melanocytes as in the normal membrane and is not columnar. The malpighian layer is well formed and supports a distinct stratum granulosum and stratum lucidum. The area immediately underlying the cyst is somewhat more hyperkeratotic than the surrounding skin.

Changes in the dermis. The most striking change in the dermis is a marked hyperplasia in the immediate vicinity of the cyst, characterized essentially by a dense proliferation of connective tissue cells of various types, chiefly fibroblasts and histiocytes. Other cell types such as mast cells and lymphocytes are scanty.

Contents of the cysts

Each mature cyst contains a single gravid female, several adult males, and a variable number of eggs and immature forms. A total count of the contents of one of the cysts showed that it held

1 female and 5 adult males, 29 eggs, and 23 immature stages (larvae and nymphs).

The large female (Figs. 11 and 12) always occupies the upper two thirds of the cyst, the other stages being restricted to the lower third (Fig. 6). Her somewhat squat-pyriform shape conforms well to that of the upper part of the chamber (considerable modifications on the generalized acarine plan adapt her to life within the cyst). The long axis of the idiosoma lies parallel to the wing membrane, with the result that the relatively small gnathosoma is directed to the side wall of the cyst. The anus, however, has rotated to a dorsal position and lies at the apex of a distinct

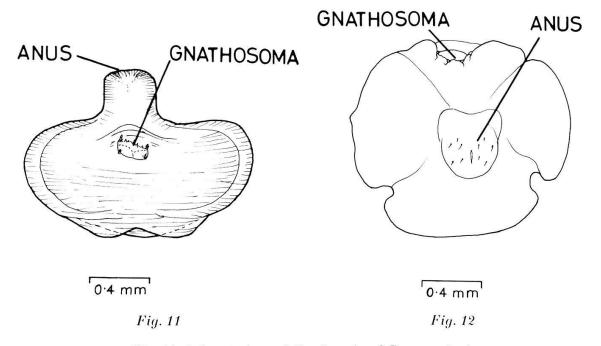


Fig. 11. A front view of the female of B. cynopteris.

Fig. 12. A drawing of the female of B. cynopteris as seen from above. This would be the appearance of the mite if it could be viewed through the cyst orifice.

prominence which can be seen through the cyst opening. In this way we were able to record the survival of a female *Bakerocoptes* cynopteris within a wing cyst in a dead bat which had been kept in a refrigerator over a week-end at a temperature of 5° C.

The other stages form a 'bed' upon which the female rests. She either lies free in the cyst cavity or burrows into the hyperkeratotic stratum corneum lining.

The systematic position of Bakerocoptes cynopteris

B. cynopteris was described by FAIN in 1962 from females, males, protonymphs, larvae and eggs removed from intradermal

cysts in alcohol-preserved specimens of Cynopteris brachyotis angulatus (Miller).

FAIN ascribed B. cynopteris to the family Teinocoptidae, but drew attention to certain peculiar features which gave it a unique position in the family. One striking feature is the neotenic character of the male; it is hexapod and devoid of sclerotised plates. Despite its neotenic appearance, however, the male possesses well developed genitalia. The extraordinarily long and stout penis differs strikingly from the much smaller and shorter intromittent organs of other described males of sarcoptiforms which have been attributed to the families Teinocoptidae and Sarcoptidae.

Bakerocoptes appears to be related to Chirobia (family Teinocoptidae), but the two genera differ in some important respects. The females in both cases are globular or subglobular, with the legs much reduced. The anus, however, is clearly centro-dorsal in position in Bakerocoptes, whereas in Chirobia it is either subterminal or postero-dorsal; moreover the structure of the anal field is different in the two genera. Unfortunately the only Chirobia male known is that of C. congolensis, but this shows no evidence of neoteny and its genital apparatus differs markedly from that of B. cynopterus; like other teinocoptids, it has a short penis. Leg IV is well developed though of smaller size, while the epimera of leg I is forked. In B. cynopteris, there is no trace of a fourth pair of legs and the epimera of leg I is unforked.

Bakerocoptes cynopteris differs so markedly from other members of the family Teinocoptidae that its retention in the Teinocoptidae does not seem justified. We therefore propose that the subfamily Bakerocoptinae of Fain should be raised to family rank and that B. cynopteris should be considered as belonging to the family Bakerocoptidae.

Discussion

While there is an extensive literature devoted to the many interesting ectoparasites of bats, the majority of papers simply describe—or redescribe—the various mites, ticks, flies, and other arthropods parasitic on these mammals. Very little has been written about the host reactions, although there are many summary references and short notes on lesions caused by ectoparasites.

Ectoparasites which do not penetrate the integument of the host to find shelter (as do, for example, bugs and nycteribiids) cannot normally be expected to produce marked reactions. But streblids, such as Ascodipteron, and mites belonging to the families

Sarcoptidae and Teinocoptidae, which are tissue-dwelling, produce changes which occasionally may be quite severe. Teinocoptids as a rule only become superficially attached to the integument (Rodhain, 1923), whereas many sarcoptids have developed more fossorial habits, some species inhabiting deep epidermal-lined burrows, which extend into the dermis (Roessler, 1932).

Most of our knowledge of the host-parasite relationships of the sarcoptiform mites of bats is based on the work of four investigators (Rodhain, 1923; Roessler, 1932; Vitzthum, 1943; and Lukoschus, 1962). Useful as they are, these studies leave much to be desired, particularly with respect to the histopathology of the lesions.

The host reaction caused by $B.\ cynopteris$ appears to be quite unlike any previously described sarcoptiform-induced reaction in Chiroptera or, for that matter, in other mammals. In general the mammalian host reaction to sarcoptid mites is characterized by a hyperkeratosis of the affected area, the mites tunnelling in a somewhat haphazard fashion within the thickened stratum corneum.

In the Chiroptera, however, the 'home' of the mite appears to assume an architectural pattern which is characteristic of the species. Roessler (1932) has shown that Notoedres roessleri produces a peculiar horn-like structure on the lip of Myotis nigricans which juts out from the surface of the skin. This horn-like structure is composed of cornified stratum corneum forming a tube, at the bottom of which the adult female mite dwells. Lukoschus (1962) has described small dome-like swellings caused by N. poppei on the wing membrane of several species of bats. These swellings are chiefly composed of a cup-like extension of the stratum corneum which almost completely encloses the gravid female, except for a break at the summit of the dome. Rodhain described a similar formation in Teinocoptes though in this case the stratum corneum forms a sleeve enclosing the anterior (attached) part of the mite; if the mite is detached from the skin, it leaves a cupule, formed from the stratum corneum. In none of these instances has the mite been described as lying in an epidermis-lined cyst in the dermis, as does Bakerocoptes.

The formation of subepidermal pouches has been previously described in other mites, but not in the Sarcoptiformes. Flynn & Jaroslow (1956) and others have given an account of the pouches formed by *P. musculi* in the skin of a mouse, while Hyland (1961) describes a very similar structure in an amphibian. Mite-containing pouches are well known in *Demodex*. The only other sarcoptiform which has been shown to form epidermal

pouches is the bird parasite Knemidokoptes pilae (Yunker & ISHAK, 1957), but these are not analogous and are only found in severe cases. Epidermal pouches are rarely formed by sarcoptids in mammals and occur only in heavy infestations.

Roessler has used the term thylacium to describe the peculiar tube-like excrescence produced by N. roessleri on Myotis nigricans. Dogiel (1964) has substituted for thylacium the term zoocoecidium, which he describes in the following terms: 'Zoocoecidia differ from ordinary swellings or capsules in the constancy of their shape and in a certain polarity, imparted to them by some structural details. Zoocoecidia are most often caused by certain ectoparasites. Settling on the surface of the hosts, these parasites produce a depression, developing into a deep pit and communicating with the outside by a more or less narrow 2 opening. Good examples of zoocoecidia are those produced by the copepod Pionodesmotes phormosomae in the sea urchin, Phormosoma uranus and by the nematode Filariodes osleri in the dog (Dogiel, 1964).

According to the strict definition of the term as given by DOGIEL, the cyst formed by B. cynopteris is a true zoocoecidium. We have sectioned numerous cysts produced by B. cynopteris and always found them to have a constant shape and polarity. Dogiel would probably not describe the tube-like structure discovered by Roess-LER in Myotis nigricans as a true zoocoecidium, and it seems wiser at this stage to restrict the term to the type of cystic formation as shown by B. cynopteris. Further study of the host reaction to Notoedres roessleri is necessary since it was not possible for Roessler, with only 2 specimens, to say if the N. roessleri 'horn' is of constant shape and polarity. Dr. A. Beck informs us that he has seen a similar host reaction in Myotis yumanensis caused by Notoedres myotis, but histological material is not available at present for study.

Zoocoecidia appear to be rare in Malaysian Chiroptera. We have examined a considerable number of bats, but have never seen anything resembling the cysts of B. cynopteris. Dr. Beck has examined over 20 000 bats in California without finding a similar structure.

Although we have inspected many specimens of Cynopterus brachyotis, we have never found an individual showing early stages of infestation by the mites. We examined one young individual carried on the mother's breast but it proved negative; in a few cases we found recently weaned bats showing well developed, ripe zoocoecidia. We therefore do not know at what stage the bats become infected. It is possible that the young becomes

² Our italics.

infested from the mother to which it clings tightly and that we have not examined enough individuals to find this. Adult-to-adult transfer is also possible. The habit of roosting communally must greatly favour exchange of ectoparasites between adult fruit-bats.

An experimental study of the host-parasite relationship should not be too difficult as the bat is very common in Singapore and can be readily trapped in mist-nets. It is easy to keep alive in the laboratory, and can be fed on a variety of fruits. The bats are easily immobilized with barbiturates or ether and can then be handled with impunity. Also since the wing membrane is thin, it can be readily transilluminated, a technique which should permit direct observations on the penetration of the host's tissues by the mites.

Since we have not been able to follow the development of the zoocoecidium from the moment of its formation, our investigation of the host reaction must be regarded as only a preliminary study of this most interesting association. We hope, however, that our study will stimulate other investigators to undertake an experimental study of the life-cycle and host-parasite relationships of *B. cynopteris*.

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Zusammenfassung

- 1. Das Verhältnis Wirt-Parasit von Bakerocoptes cynopteris, einer parasitischen sarcoptiformen Milbe, und dem Flughund Cynopterus brachyotis wird histologisch untersucht.
- 2. B. cynopteris verursacht Cysten von 1,5-2 mm Durchmesser in der Flügelmembrane des Wirtes. Jede reife Cyste enthält ein trächtiges Weibchen, mehrere Männchen und verschiedene unreife Stadien.

- 3. Eine Präparationstechnik für histologische Schnitte der Cysten wird beschrieben.
- 4. Histologisch besteht jede Cyste aus einer geräumigen, von Epithel umgebenen Kammer, welche zwischen der oberen und unteren Fläche des Flügels liegt. Der Hohlraum der Cyste öffnet sich nach außen durch eine zentral gelegene kleine Öffnung. Das Epithel, das die Innenwand der Kammer umschließt, wie auch dasjenige, das über der Cyste liegt, ist deutlich acanthotisch. Hyperkeratosis fehlt im allgemeinen mit Ausnahme im Hals der Cyste.
- 5. Die Unterfamilie Bakerocoptinae Fain wird in den Rang einer Familie Bakeroptidae erhoben.

Résumé

- 1º Une étude histologique concernant les relations hôte-parasite entre l'acarien sarcoptiforme Bakerocoptes cynopteris et le chiroptère Cynopterus brachyotis a été entreprise.
- 2º B. cynopteris produit des kystes mesurant de 1,5 à 2 mm. Ceux-ci sont localisés dans la membrane alaire de l'hôte. Chaque kyste mûr contient une femelle gravide, plusieurs mâles et des stades en développement.
 - 3º On décrit une technique servant à la préparation histologique des kystes.
- 4º Du point de vue histologique, chaque kyste consiste en une chambre spacieuse, limitée par un épithélium, comprise entre les deux surfaces de l'aile. La cavité du kyste s'ouvre à l'extérieur par un petit orifice central. L'épithélium tapissant la paroi de la cavité et celui entourant le kyste sont distinctement acanthotiques. Une hyperkératite est généralement absente excepté dans le goulot du kyste.
- 5º La sous-famille Bakerocoptinae Fain 1962 est élevée au rang de famille Bakerocoptidae.