INTRODUCTION
Mesothelae have not been spared by predators, parasites and diseases. Besides ravaging animals like skinks and centipedes which regularly come across those soil dwelling spiders, there are reports on parasitic laelapid mites like Liunghia bristowei Finnegan, 1933 living on Liphistius malayanus Abraham, 1923 and there are parasitic flies and wasps (Sawaguti and Ozi 1937; Bristowe 1976; Schwendinger 1990) which attack Mesothelae along with other spiders. Mesothelae are also subject to parasitation by mermithid nematodes (pers. obs.).

Infections found in some species of Mesothelae are compared to those in other trapdoor spiders of the genus Latouchia (Mygalomorphae, Ctenizidae) which attack Mesothelae along with other spiders. Mesothelae are also subject to parasitation by mermithid nematodes (pers. obs.).

MATERIAL AND METHODS
Specimens of Mesothelae and Ctenizidae (genus Latouchia) collected in Kyushu, in Okinawa and in Malaysia were brought to the laboratory and kept there for ethological and morphological studies. The determination of mesothele spiders is according to Haupt (1983). Specimens of Latouchia were only determined to the rank of genus which is sufficient for the present work. The fungus Nomuraea atypicola (Yasuda, 1915) Samson, 1974 (syn. Isaria atypicola, Cordyceps atypicola) (Deuteromycotina: Hyphomycetes) was identified according to Kobayasi (1982). Rickettsiales could only be identified as an order, according to the electron microscopic appearance of the cells.

As in several species the collected material was quite numerous, it represented a good occasion to study the prevalence of different pathogens. The present study was carried out on living spiders, except for Nomuraea infections in which the perithecial stroma grows out from the dead spider and opens the trapdoor from inside the burrow, thus indicating the infection.

Trapdoor spiders live inside their burrows in moist soil, therefore the observer easily misses the date when a specimen dies. In the
moist environment dead spiders may become infected secondarily by fungi from the soil: such cases were not studied in the present paper.

For light microscopic study, the opisthosoma of infected specimens was submersed in Bouin’s fluid and embedded in paraplast. Sections were stained with Haematein, Azan or Haematoxylin/Chromotrop after Dobell. Procedures for electron microscopy were as described previously (Haupt 1996), semi-thin sections (1 µm) were stained with Kristallviolett (Merck). Specimens of Nomuraea atypicola were deposited in the Botanical Museum, Berlin.

RESULTS

Histological and electron microscopic results
In Mesothelae, rickettsial infection is very obvious in the intermediate cells of the hepatopancreas (Fig. 1), while neighbouring cells of the midgut remain uninfected. The intermediate tissue is known to function as storage organ

Fig. 1. Electron microscopic section through the opisthosoma of Liphistius malayanus cameroni with intestinal cells (I) and rickettsiae in intermediate cell (R). P: vacuoles containing protein, V: vacuoles. Scale 10 µm.

Fig. 2. Perithecial stroma of Nomuraea atypicola emerging from the burrow of Latouchia sp. (Ctenizidae) by opening the trapdoor from inside. Scale 2 mm.
analogous to the fat body of insects. Mainly lipids and glycogen are found in these cells (Ludwig & Alberti 1988), but the histological study also reveals numerous vesicles containing protein.

The first rickettsial stages to be detected are narrow and electron dense. Apparently, they grow inside the host cells to form larger, less electron dense cells. This stage may or does undergo binary fission thus multiplying the parasitic population in the host cell. Finally, all intermediate cells in the hepatopancreas are filled with nothing but rickettsial cells: all organelles and even the nuclei have disappeared. Apparently, the rickettsiae use up the storage products for their own purpose, and moulting becomes impossible for the spider.

These later stages of infection can be recognized easily by the whitish opaque opisthosoma of host spiders, which is typical for rickettsial infections. Generally, this stalk, the perithecial stroma, grows along the burrow, opens the trapdoor from inside, and reaches a height of 20 to 30 millimeters. In the upper part it bears numerous purple ascospores (Fig. 2), which now, outside the spider’s burrow, are subject to aerial dispersal.

Instead, during 20 years of breeding Mesothelae, out of 549 specimens taken from the field not a single one was found to be infected by *Nomuraea atypicola*.

### Infection rates

There are regular infections by Rickettsiales in Mesothelae (Table 1) found in Kyushu, Ryukyu, or Malaysia. The infection rate must be considered low, as far as most localities are concerned. Nevertheless, when comparing infection rates of the same species from different localities, there were striking differences: Rickettsial infection rates of *Ryuthela nishihirai nishihirai* were almost 6% (n = 52) in Suyeyoshi, Naha, Okinawa, but almost negligible in Ryutan, Naha, Okinawa. Both places, only a few kilometers from each other, are or were residues of rather natural habitats with similar soil conditions, but the locality in Suyeyoshi is situated in a valley of a brooklet, while Ryutan is close to a hill top, more exposed to wind and this place appears much drier.

The other group of trapdoor spiders, living in the same habitats, belongs to the genus *Latouchia* (Ctenizidae). Among them, one may regularly observe infections by the fungus *Nomuraea atypicola*. Its hyphae grow through the whole body of the spider and finally, within a few hours, form a long stalk with conidium production. Generally, this stalk, the perithecial stroma, grows along the burrow, opens the trapdoor from inside, and reaches a height of 20 to 30 millimeters. In the upper part it bears numerous purple ascospores (Fig. 2), which now, outside the spider’s burrow, are subject to aerial dispersal.

<table>
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<tr>
<th>Table 1. Number of specimens of Mesothelae and <em>Latouchia</em> (Ctenizidae) collected (N) compared to those infected by different pathogens.</th>
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<tbody>
<tr>
<td><strong>Heptathela kimurai kimurai</strong> (Kishida, 1920)</td>
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<tr>
<td>Heptathela kimurai yanbaruensis Haupt, 1983</td>
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<tr>
<td>Ryuthela nishihirai nishihirai (Haupt, 1979)</td>
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<tr>
<td>Liphistius malayanus cameroni Haupt, 1983</td>
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<td>Latouchia sp.</td>
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So far, no experimental infection of Mesothelae along with food has been successful.
DISCUSSION
Many fungal infections have been described in connection with spiders (Evans & Samson 1987), but unfortunately, in many cases either the spider or the fungus remained undetermined. The hyphomycete fungus *Nomuraea* certainly occurs in a wide range of soil dwelling mygalomorph spiders (for literature, see Coyle et al. 1990). In North America a broad range of spider species could be infected experimentally by an isolate of *Nomuraea* from ascospores originating from an infected Brazilian trapdoor spider. By applying a conidial suspension containing a detergent, 20 out of 27 spider species were successfully infected (Greenstone et al. 1987). In Okinawa, on the other hand, infectious diseases in trapdoor spiders seem to be clearly linked to different families: while *Latouchia* (Ctenizidae) specimens were found to be infected by *Nomuraea atypicola*, rickettsial infections were limited to Mesotheleae. So far, the experimental infection of Ryukyuan mesothele spiders by *Nomuraea* (without using detergents) turned out to be impossible.

Among Rickettsiales, the genus pathogenic in arthropods has been named *Rickettsiella*. Very similar pathogens as in Mesotheleae have been reported from intermediate (interstitial) cells of a Japanese funnel web spider (*Paracelotes lucutusus*) (Osaki 1973) and from a linyphiid spider (Suhr 1995). These pathogens are comparable to infections described from the hepatopancreatic caeca in a buthid scorpion (Morrel 1976).

In acarids (Reinhardt et al. 1972), insects, diplopods (Schlueter & Seifert 1985) and woodlice (Vago et al. 1970) the occurrence of similar pathogens is quite frequent, and they have been reported as intracellular parasites from different tissues, such as labial glands (*Lepisma saccharina*, pers. obs.), Malpighian tubes (Schlueter & Seifert 1985) and adipous tissue (Götz 1972; Vago et al. 1970). In the case of woodlice, there is strong evidence that specimens from moist and periodically wet habitats are most likely to be infected (Federici 1984), which is in line with our observations on *Ryuthela nishihirai*.

The wide distribution does not automatically allow any conclusion on specific identity. At present it seems doubtful whether pathogenic Rickettsiales represent only one species. Transmission of Rickettsiales from a scorpion to other arachnids, even to members of other scorpion families, turned out to be impossible (Morrel 1978). Our studies on infections of trapdoor spiders point to the same direction. The redefinition of the group based on serological data has been strongly suggested previously (Louis et al. 1977).

In general, the infection rate with Rickettsiales is low. A 5% infection rate in scorpions (Morrel 1976) is well in line with our results. Such low prevalence of infection may also be the reason that early stages of infections are hardly found in the intermediate cells of free-living Mesotheleae; only the late stage of infection is obvious.

There are interesting reports on Rickettsiales and unidentified virus particles occurring in acarid ovaries (Lewis 1979), in insect sperm, spider coenosperma, and mite spermatozoa (Afzelius et al. 1989). Apparently, still other paths of infection are used beside food, and *Wolbachia pipiens* is known to commonly infect ovary cells, thus being transmitted in a vertical manner (Bourtzis & Braig 1999).

REFERENCES


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