Demonex flagellurus sp. n.

(ACARI: DEMODICIDAE) FROM THE PREPUTIAL AND CLITORAL GLANDS OF THE HOUSE MOUSE, MUS MUSCULUS L.

V. RUKVA

Institute of Parasitology, Czechoslovak Academy of Sciences, České Budějovice

Abstract. An extremely large new demodicid, Demonex flagellurus sp. n., is described in all stages of its life cycle from the house mouse, Mus musculus L. Its habitat, the preputial and clitoral glands, is reported for the first time for demodicid mites. Adaptation of habitat, host transfer, and the potential of this new species as a useful model in experimental studies on demodicid biology are discussed.

Hirst (1917, 1919) referred to a single demodicid species—Demonex arcicolas var. musculi Oudemans, 1898—in the house mouse. However, his data and figures suggest the existence of (at least) two Demonex species in this host. The presence of an unnamed Demonex sp. on the mucosa on the tongue of the laboratory mouse was reported by Tuzdil (1957).

A previously unreported Demonex species was found by us in the preputial and clitoral glands of the house mouse. It is described below as a new species, with some information on its habitat, prevalence, populations and pathogenicity.

MATERIAL AND METHODS

A total of 434 (198 male, 236 female) house mice from wild populations in South Bohemia were examined for demodicids. Forty-seven mice were examined in 9 body areas (oral cavity, vibrissae area, eyelids, vertex, ear, dorsum of trunk, genital area, anus, tail); 387 mice were examined in genital areas only. Over 1,000 mite specimens were obtained by squeezing out the host's penis or clitoral elevations and the underlying tissues with washmaker's forceps, and/or excising these organs and dissecting them with dentist's scissors. The obtained oily secretions containing mites were diluted and cleared with Hoyer's medium, and mounted into permanent mounts. Morphological characters of mites were studied with phase-contrast immersion optics. Histological data were obtained from samples of genital areas of 17 mice fixed in 10% neutral formalin, paraflin-embedded, sectioned at 8 to 10 μm and stained with haematoxylin-eosin. Anatomical structures of mice were interpreted after Cook (1965), Green (1966), and Guze et al. (1982). Phases of sexual development of mice (juveniles, subadults, adults) were determined from gonads using routine mammalian criteria.

RESULTS

Demonex flagellurus sp. n.

An extremely large species, strongly elongate in posterior part of body in both sexes and all stages of the life cycle.

Male (holotype) (Figs. 1E; 2A, B; 3A, C, D, E). Body length 787 μm with opistho-
soma comprising approximately seven eighths of this value. Other measurements are given in Table 1.

Gnathosoma slightly wider than long. Palpal tarsus with two 2-tined and one simple spines. Supracoaxal spines small, approximately 3 mm long, straight, slightly conical with blunt apices, dorso-medio-posteriorly oriented. Pharyngeal bulb horsehoe-shaped, open posteriorly. Subgnathosomal setae anterolateral to anterior edge of pharyngeal bulb. (Figs. 2A, B; 3C, E).

Table 1. Means and standard deviations of 20 specimens of each stage and sex of *Demodex fagellarus* sp. n. (all measurements in μm)

<table>
<thead>
<tr>
<th></th>
<th>Male</th>
<th>Female</th>
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<tbody>
<tr>
<td></td>
<td>Length</td>
<td>Width</td>
</tr>
<tr>
<td>Gnathosoma</td>
<td>24.0 ± 2.0</td>
<td>25.4 ± 1.3</td>
</tr>
<tr>
<td>Podosoma</td>
<td>93.8 ± 4.4</td>
<td>61.5 ± 4.5</td>
</tr>
<tr>
<td>Opisthosoma</td>
<td>571.2 ± 76.2</td>
<td>49.7 ± 13.1</td>
</tr>
<tr>
<td>Total length</td>
<td>689.0 ± 76.3</td>
<td>476.3 ± 13.2</td>
</tr>
<tr>
<td>Aedeagus</td>
<td>48.3 ± 11.1</td>
<td>—</td>
</tr>
<tr>
<td>Vulva</td>
<td>—</td>
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Aedeagus slender and long (49 mm), swollen at base. Central orifice mid-dorsally between anterior pair of dorsal podosomal tubercles, a simple slit flanked by paired elevated sclerites antero-laterally and posteriorly (Fig. 3A).

Podosoma comprises approximately one eighth of total body length. Four pairs of legs evenly spaced along podosoma; approximately four fifths of 3-segmented free parts of strong legs are visible from dorsal aspect. Sutures of epimeral plates indistinct towards the midline. Movable segments 2 with minute posteroventral spurs. Each leg with two unequal claws; each of them bifid distally and with a spur projecting from shaft. Legs I and II with a balliform solenidion near the claws. Dorsum of podosoma with striae at the level of anterior half of coxal plates I, transverse smooth area with elevated lateral edges at the level of posterior half of coxal plates I, fingerprint-like inscription of striae at the level of coxal plates II—III, and transverse striation at the level of epimeral plates IV. In corners of the dorsum smooth area four dorsal podosomal tubercles are arranged to form a trapezoid with parallel sides 42 μm and 33 μm long, respectively. Tubercles not very prominent, each with a minute seta seen as a dark point (Figs. 2A, B; 3A, D).

Opisthosoma extremely long and slender, tapering to whip-like apex. Annulation regular, narrow, and faint. Anterior part of opisthosoma with a hemispherical to conical ventral protuberance without annulation. Opisthosomal organ absent.

Female (allootype) (Figs. 1 F; 2 C, D; 3 B): Shorter than male; body length 430 μm with opisthosoma comprising three-fourths of this value.

Gnathosoma and relevant structures similar to male; width of gnathosoma larger, supracoaxal spines shorter than in male, oriented upwards.

Podosoma comprises approximately one fifth of total body length. Legs shorter than in male; their free parts one-third to two-thirds visible from dorsal aspect. Epimeral sutures distinct, meeting at midline. Other structures of legs identical with male. Dorsum of podosoma smooth at the level of epimeral plates I—III, with transverse striation over epimeral plates IV. Two pairs of shallow circular impressions on dorsum at the level of anterior borders of epimeral plates II and III. Dorsal podosomal tubercles absent. (Fig. 2 C, D).

Opisthosoma shorter than in male, tapering to acute apex more abruptly than in male. Dorsum near base of opisthosoma with a pair of minute tubercles. No ventral protuberance. Other opisthosomal structures identical with male.

Vulva 12 μm long, a longitudinal slit opening behind epimeral plates IV (Fig. 3 B).

Ovum (Fig. 1 A). Elongate drop-shaped, tapering to whip-like apex posteriorly, on the average 271.4 μm long and 25.6 μm wide. No operculum groove observed.

Larva (Fig. 1 B). General body shape similar to ovum; three legged-stage, on the average 363.4 μm long. Palpal tarsus terminally with two spines; of these ventral spine 2-tined, dorsal spine simple; medio-proximally a thin simple spine. Pharyngeal bulb similar to adults. Subgnathosomal setae absent. Each leg with a single 4-pointed claw. Integument without annulation. Solenidia and epimeral setae not observed.

Fig. 1. *Demodex fagellarus* sp. n., body outlines. A — ovum; B — larva; C — protonymph; D — nympha; E — male, arrow — ventral protuberance; F — female. Relative sizes.
Protonymph (Fig. 1 C). Similar to larva except for larger size (on the average 661.9 μm long) and end of each leg with two 4-pointed claws. Gnathosoma and somatic structures, integument, solenidia, and epimeral scutes as in larva.

Nymph (Fig. 1 D). Similar to protonymph except for four legs and larger size (on the average 1010.2 μm long, the longest specimen measuring 1184 μm). Gnathosoma and somatic structures similar to protonymph. Each leg with two 4-pointed claws. Solenidia not observed. Three pairs of epimeral scutes between legs II-IV; all scutes oval, pressed onto surface of integument, indistinct (not observed in all specimens). Integument without annulation.

Differential diagnosis. Demodes flagelliferus most closely resembles two demodicids from Neotropical bats: D. longissimus Desch, Nutting et Lukoschus, 1972 from Carolia persicillata (L., 1758) and D. melanopteri Lukoschus, Jongeman et Nutting, 1972 from Eptesicus melanopterus (Jentink, 1904). The characters these three forms have in common are: a great total length (D. longissimus female 790.0 μm, male 737.7 μm; D. melanopteri female 425 μm, male 419 μm) and an extremely elongate opisthosoma (comprising three-fifths to four-fifths of total body length in D. longissimus and D. melanopteri).

The following characters serve to differentiate D. flagelliferus: 1. Male longer than female (vs. female slightly longer than male in D. longissimus; female as long as male in D. melanopteri); 2. Posterior of opisthosoma extremely narrow, with very thin acute apex; apex of opisthosoma whip-like in male, larva, protonymph, and nymph (vs. posterior of opisthosoma broader, with blunt apex in all stages of D. longissimus and adults of D. melanopteri; no data on the shape of opisthosoma apices have been published for larva, protonymph and nymph of D. melanopteri); 3. Ovum non-oviparous, elongate, drop-shaped with whip-like apex (vs. ovum operculate in D. longissimus and D. melanopteri; ovum extremely long and snake-like in D. longissimus, drop-like and pointed abruptly on apex in D. melanopteri); 4. Larva, protonymph and nymph without conspicuous holdfast structures (vs. elongate, claw-like legs III modified to holdfast organs in larva, protonymph and nymph in D. longissimus).

Besides D. flagelliferus several other Demodex species parasitize the house mouse (Hirst 1917, 1919, Tüdül 1915; our material to be described later). D. flagelliferus is not likely to be confused with any one of them, as they are shorter, with distinctly broader and less elongate opisthosomata. Hirst (1919) gives the following measurements for his form(s): total length of female 113–170 μm, male 120–154 μm, deutonymph 235 μm. Demodex sp. from the oral cavity of the laboratory mouse is 400 μm long (Tüdül 1915). An undescribed Demodex sp. from the eyelids, the second longest demodicid collected from the house mouse by us, is 296–333 μm long (5 females). Additional differences between D. flagelliferus and these synnathous demodicids are indicated in other standard taxonomic characters.

Host and locality: Mus musculus musculus L., 1758, the house mouse; wild populations in South Bohemia, Czechoslovakia.

Type material (holotype, allotype; para- and sub-para material) from a host collected by Dr. M. Vlček at České Budějovice on Oct. 31, 1962. Types are deposited in the collections at the Institute of Parasitology, Czechoslovak Academy of Sciences, České Budějovice. Holotype No PaP ČSAV 1933 and allotype No PaP ČSAV 1934. Some para- and sub-para material is deposited in the collections at the University of Massachusetts, Amherst, USA.

Prevalence, habitat, populations, pathogenicity. A total of 34 (9 males, 16 females) of 434 hosts examined (8.8%) were found to be infested with D. flagelliferus. Of the infested hosts two (females) were juvenile, six (females and males) were subadult, and sixteen (females and males) were adult.
The mites were detected only in hosts' genital areas. The ease with which the mites contained in profuse oily secretions were squeezed out from near the apices of the penile or clitorial elevations evidenced their site in large glandular structures opening to the preputial or clitoral glands. Four most heavily infested hosts (2 males, 2 females) of those examined by squeezing out their genital elevations yielded 74 to 250 mites (all stages). A total of 531 mites collected from these four hosts were represented in the following proportions: males 71.1%, females 40.1%, ova 16.5%, larva 5.5%, protonymphs 6.2%, nymphs 24.3%. Considering the deep localization of early stages of the life cycle (see below) and the examination technique, these figures may underrate the preimaginal stages.

**DISCUSSION**

Our observations suggest that *D. flagellarus* is restricted in its distribution to the preputial and clitoral glands. To our knowledge, this is the first record for this habitat for demodicid mites.

The preputial and clitoral glands, homologous organs of male and female mice, are large glands of sebaceous type. Green (1966) describes them to be made up of groups of flat polyhedral cells with pale-staining nuclei, surrounded by connective tissue capsules. In an actively secreting cell, the major portion is filled with an oily secretion, and when the nucleus degenerates the dead secretion-packed cell is excreted. The ducts of the glands are lined with stratified squamous epithelium; they open into the preputial cavity or in the lateral wall of the clitoral fossa.

Similar habitats of demodicid mites are the Meibomian glands in the eyelids of mammals. They are large multi-alveolar sebaceous glands producing a paste-like sebum which is excreted through central ducts lined with stratified squamous epithelium; they open on margins of the eyelids. The main ecological characteristics of the Meibomian glands are the tubular shape of the ducts and the constant sebum flow tending to expulse the mites especially in the inactive stage of ovum and during the inactive molting intervals between the developmental stages.

Various holdfast adaptations of demodicids to match these conditions have been described, e.g., T-shaped ova in *Demodex gapperi* (Nuttall et al. 1968), and *D. huttereri* (Mertens et al. 1983), T-shaped ova in *D. molestus* (Desch et al. 1972) and *D. peromysci* (Lombert et al. 1983), ova with bulbous anterior and elongate ends in *D. longissimus* (Desch et al. 1972) and *D. melanopleri* (Lukoschus et al. 1973). In the larval and nymphal stages, dorsal finger-like protrusions of podsoma have been described in *D. gapperi* (Nuttall et al. 1971), *D. peromysci* (Lombert et al. 1983), and *D. huttereri* (Lombert et al. 1983), dorsal only; finger-like protrusions in *D. molestus* (larvae only; Desch et al. 1972), elongate claw-like legs III in *D. longissimus* and *D. molestus* (Desch et al., 1972), exceptional broadening of podsoma in *D. bicoedatus* (Kniest and Lukoschus 1981), (Nuttall et al. 1980), prolonged supracloacal spines in *D. marsupialis* (Nuttall et al. 1980), long legs with broad multicellular claws in *D. laticeps* (Lukoschus and Jongman 1974), *D. melanopleri* (Lukoschus et al. 1973), *D. bicoedatus* (Kniest and Lukoschus 1981), *D. huttereri* (Mertens et al. 1983), and *D. intermedicus* (Lukoschus et al. 1984). Some demodicids inhabiting the Meibomian complex are very large species with remarkably long opisthosoma and (in adults) prominent strong legs (*D. longissimus*, *D. molestus*, *D. melanopleri*).

Desch et al. (1979), Lukoschus and Jongman (1974), Lombert et al. (1983) and Lukoschus et al. (1984) discuss the adaptive features in demodicids inhabiting the Meibomian ducts with respect to duct dimensions. They note that the most pronounced adaptations are shown in species inhabiting the wide ducts, *D. molestus* (duct diameter 220 µm) and *D. longissimus* (duct diameter 120 µm). As the clitorial ducts containing *D. flagellarus* were observed to be considerably wider (up to about 700 µm), it is interesting that—compared with *D. molestus* and *D. longissimus*—the series of adaptive features in *D. flagellarus* is rather limited and achieved through relatively minor modifications:

1. Extremely large body size and elongation of opisthosoma. *D. flagellarus* is the largest known demodicid, its nymphal stage being by approximately 100 µm longer than the longest stage of *D. longissimus*.

2. Elongate shape of large ovum.

3. Prominent strong legs with long claws in adult mites (especially males).
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Received 2 May 1984.

Acknowledgement. The author wishes to thank Dr. W. B. Nutter of the University of Massachusetts, Amherst, for suggesting D. flagellatus to be a helicerno underdescribed mite species. Thanks are also due to the author’s colleagues of the Institute of Parasitology, Dr. J. Vitovec for revising some histological sections and reading the part of the manuscript dealing with pathogenetics. Mrs. E. Wilkova for supplying host material, and Mrs. V. Richturova for expert technical assistance. The author is grateful to Dr. W. D. Gude of the Oak Ridge National Laboratory, Oak Ridge who kindly provided a copy of his Histological Atlas of the Laboratory Mouse.

DEMODEX FLAGELLATUS SP. N. (ACARI: DEMODICIDAE) FROM PREPUCTIONSAL AND CLITORAL MICE DOMOVY MYSI MUS MUSCULUS L.

B. Buicza

Rézuma. Datumo izobrazenie novogo viza demeljnic Demez flagellatus sp. n., vo vseh stadih ego izjavnogo obidnaja, o domovy mysli mus musculus L. Esto mestobabo — pravylalna i klitoralny mely — pridavajsa novym do demeljnice. Obussiayta adaptatsia i mestobabo, preredaha sredazh zeshem i vozvazhka dostrika, to ego novogo viza v izobredesti i eksperimentalih rabotih po biologii demeljnic.
Figs. 1, 2. Habitat of *Demodex flagellatus* in the preputial glands of *Mus musculus*. Fig. 1. Section through the penis elevation to show the penis (p) in the preputial cavity, and the preputial gland with mites. Only largest groups of mites (m) indicated (×23). Fig. 2. Preputial gland containing mites (m) (×100).

Figs. 1, 2. Habitat of *Demodex flagellatus* in the clitoral glands of *Mus musculus*. Fig. 1. Section through the clitoral elevation to show erectile tissue surrounding other structures of the clitoris (et), and the clitoral glands (cg) with the ducts containing mites (dm) (×40). Fig. 2. Duct of the clitoral gland containing mites (m) (×165).