genus *Clemmys*, it is our opinion that *E. megalostiedai* may also be separated from the existing species reported from turtles on the basis of host specificity. The specific epithet given to this new parasite is of Greek derivation and means "large Stieda body."

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Description and Bionomics of *Octomyomermis troglodytis* sp. n. (Nematoda: Mermithidae) Parasitizing the Western Treehole Mosquito *Aedes sierrensis* (Ludlow) (Diptera: Culicidae)

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ABSTRACT: *Octomyomermis troglodytis* sp. n. (Nematoda: Mermithidae) is described from the larvae, pupae, and adults of the western treehole mosquito, *Aedes sierrensis* (Ludlow), in Marin County, California. The species possesses a barrel-shaped vagina, eight longitudinal chords, six cephalic papillae, two short separate spicules, and a cuticle without cross fibers. Specific characters include a slightly ventrally displaced mouth, an egg diameter approximately ½ the body width, and the pharyngeal tube approximately ¾ of the total body length. The adult tails never bear a terminal appendage. Parasitic development lasts from 20–22 days at 20 C and the adults remain in the moist rotted organic matter in the bottom of the treehole for most of the year. Infection occurs in March, April, and May and reached 38% in one treehole. This species could probably be mass-produced and used for biological control against *A. sierrensis* and possibly other treehole mosquitoes.

The western treehole mosquito, *Aedes sierrensis* (Ludlow), breeds in the winter and spring months in northern California. The adults feed on humans when natural hosts are not available and also serve as the intermediate host of *Setaria yehi* Dessert, a filarial parasite of deer in California. The nature of the larval habitat makes control of *A. sierrensis* difficult and a survey of its natural enemies revealed a mermithid nematode parasite (Sanders, 1972).

There are over 75 published reports of mosquitoes being attacked by mermithid nematodes; however, this is only the third account of a mermithid parasitizing mosquitoes in treeholes. Muspratt (1945) reported a mermithid attacking *Aedes* and *Culex* larvae in treeholes in Northern Rhodesia and Petersen and Willis (1969) cited a mermithid infecting *Orthopodomyia signifera* in a treehole in Louisiana. Neither of these nematodes was described or studied in detail.

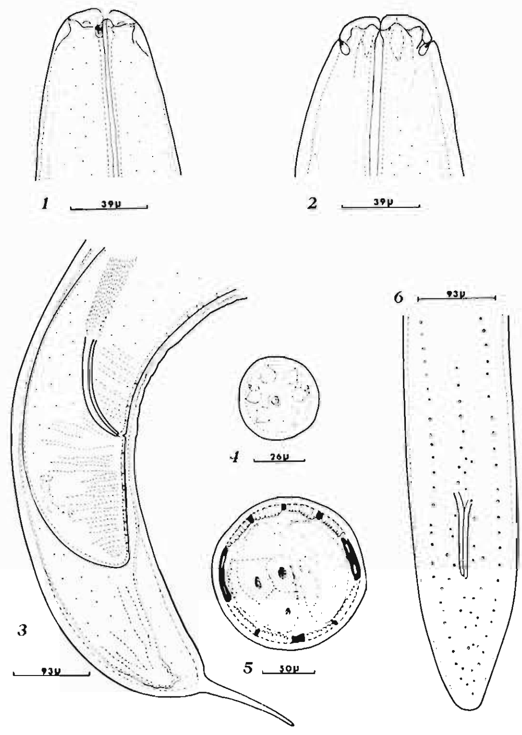
The present account describes the mermithid from *A. sierrensis* and discusses its bionomics.

Materials and Methods

Treeholes in the coast live oak, *Quercus agrifolia* Nee, and California bay, *Umbellularia californica* (H. & A.), were sampled for infected larvae and pupae of *Aedes sierrensis* (Ludlow). Out of 18 treeholes sampled, only one in *Q. agrifolia* yielded mermithid parasitized stages of *A. sierrensis*. This treehole, which was 60 cm above the ground, held approximately 23 liters of water and had an east-southeast exposure. The treehole water consistently gave a pH reading slightly above 8.

Parasitized mosquitoes were maintained at 15–20 C in sterile treehole water with a pinch of Brewer's yeast until the nematodes emerged. The postparasitic juvenile nematodes were transferred to a small petri dish with a layer of sand in the bottom and held at 20–25 C. Mermithids were directly collected from the organic matter in the bottom of the treehole by carefully sorting through samples of debris removed by hand.

Adult mermithids were fixed in TAF (triethanolamine, formalin, and water) and processed to glycerin. Histological sections for determining the number of muscle fields and hypodermal chords were made with a freeze-microtome (cryostat) and sections were cut at 6 μ and stained with a 0.025% aqueous solution of methylene blue. Morphological studies on the parasitic juveniles were made on living material stained with a 0.05% aqueous solution of New blue R.



Figures 1–6. *Octomyomermis troglodytis* sp. n. Lateral view of adult male head. 2. Ventral view of adult female head. 3. Lateral view of adult male tail with inner thin cuticle and outer thick cuticle with tail appendage of the postparasitic juvenile. 4. "En face" view of adult male. 5. Midbody cross section of adult female. 6. Ventral view of male tail with genital papillae.

Results

The mermithid nematodes found parasitizing *Aedes sierrensis* were a new species and are described below in the genus *Octomyomermis* Johnson. In the quantitative portion of the description, the first figure is the mean of observations made and the range is given in parentheses. All measurements are in microns unless otherwise specified.

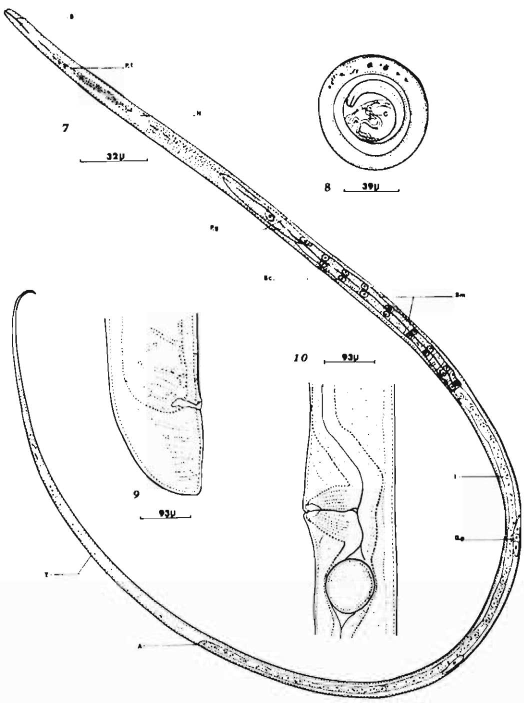
Octomyomermis troglodytis sp. n.
Mermithidae Braun, 1883.
Octomyomermis Johnson, 1963

ADULTS: Cross fibers lacking; eight hypodermal chords at midbody; six cephalic papillae in one crown; no labial papillae; mouth slightly displaced ventrally; amphids medium in size, without a commissure; tail bluntly rounded, without a mucron; pharyngeal tube extending ¾ of body length.

FEMALE (Figs. 2, 5, 9, 10) (n = 10): Length 12.6 (10.2–15.0) mm; greatest width 158 (120–165); head to nerve ring 236 (193–308); head to amphidial opening 10 (7–20); length amphidial pouch 4 (3–7); diameter amphidial opening 1.4 (0.6–2.6); distance posterior end to end of trophosome 99 (31–149); vagina barrel-shaped, length 83 (79–95); % vulva 57 (54–64); vulva protruding slightly, vulvar flap absent; egg diameter 81 (78–93); tail bluntly rounded.

All females examined possessed a rudimentary anal opening and other evidence of maleness in the tail region such as weak copulatory muscles and abortive genital papillae. Such features are characteristic of intersexes in the Mermithidae.

MALE (Figs. 1, 3, 4, 6) (n = 8): Length 8.2 (7.2–9.1) mm; greatest width 117 (100–140); head to nerve ring 211 (193–254); head



Figures 7–10. *Octomyomermis troglodytis* sp. n. Lateral view of preparasitic juvenile. S, stylet; P.t., pharyngeal tube; N, nerve ring; P.g., penetration glands; Sm, stichosome; Sc, stichocytes; I, intestine; A, anus; T, tail. 8. Egg containing a preparasitic juvenile ready to hatch; C, molted cuticle. 9. Lateral view of adult female tail. 10. Lateral view of vulva and vagina of adult female.

to amphidial opening 9 (7–13); length amphidial pouch 5 (4–7); diameter of amphidial opening 1 (1–3); length of tail 145 (130–162); width of anus 111 (100–126); spicules short, paired, parallel for most of their length, equal or subequal; length 104 (83–130); width 7 (6–9); tail blunt; genital papillae arranged in three broken rows, with the middle row becoming diffuse around the anus; each row contains from 19–23 preanal papillae and 10–15 postanal papillae.

Postparasitic juvenile (Fig. 3): Leaves the host and molts twice before reaching the adult stage; length of tail appendage 106 (91–109).

Preparasitic juvenile (Figs. 7, 8): Length (620–650). This stage contains a well-developed stylet, a pharyngeal tube, nerve ring, stichosome with 16 stichocytes, a pair of penetration glands, intestine, gonad primordium, anus, and elongate tail. The anterior portion from the head to the base of the stichosome is roughly ⅓ of the total body length; the intestine makes up slightly more than ⅓ and the tail slightly less than ⅓ of the total length. The gonad primordium is located behind the junction of the pharynx and intestine. The alimentary tract is a single unit in this stage and separates only after the nematode begins its parasitic development. The thin-walled intestine lacks a lumen and is filled with granules. The anus is faint.

Host: Body cavity of the larvae, pupae, and adults of *Aedes sierrensis* (Ludlow) (Diptera: Culicidae).

Types: Holotype (♀) and allotype (♂) deposited in the nematology collection at the University of California, Davis.

Type location: Treehole in *Quercus agrifolia* Nee near Novato, Marin County, California.

Diagnosis: A small to medium-sized mermithid with a slightly ventrally displaced mouth, a barrel-shaped vagina, eight longitudinal chords, six cephalic papillae, two short separate spicules, and a cuticle without cross fibers.

The only other species in this genus is *O. itascensis*, which Johnson described from a chironomid. However, this nematode is considerably larger than *O. troglodytis*, possesses a terminal mouth, egg diameter approximately 1/7 of the body width, the pharyngeal tube extending approximately ¼ of the total body length, and both sexes with a rounded tail sometimes with a small terminal appendage. *O. troglodytis* possesses a slightly ventral mouth, egg diameter approximately ½ the body width, the pharyngeal tube extending approximately ¾ of the total body length, and the adult tails never with a terminal appendage. The hosts of these two species belong to different families of Diptera.

The diagnosis of *Octomyomermis* presented by Johnson (1963) is accepted here. The diagnosis given by Nickle (1972) is considered incomplete since he neglected to mention the number of cephalic papillae, the absence of cross fibers, or the number of hypodermal chords. Nickle used the nipplelike tip of the tails in both sexes as a generic character; however, Johnson stated that this character was only sometimes present.

Rubtsov (1968) recently described *Capitomermis crassiderma* which possesses the basic characters of the genus *Octomyomermis*. Members of this genus possess a thick cephalic layer of cuticle, posteriorly placed amphids, few genital papillae, and pointed tails in both sexes. All the above characters separate this species from *O. troglodytis*.

Bionomics

HOST: The western treehole mosquito deposits its eggs on the moist inner surface of rot holes above the water line in spring and summer. The fall rains wash the eggs into the filling treeholes and hatching occurs under favorable conditions. The second-, third-, and fourth-instar larvae may remain in the treehole for months, but pupation generally occurs in late winter and spring. The adults may be collected in all months except November and December and the female is long-lived and may take several blood meals before the deposition of each egg batch. This mosquito is usually a univoltine species; however, late spring rains may initiate a second generation.

PARASITE: *O. troglodytis* generally emerged from fourth-stage mosquito larvae, although pupae and adults were also infected. Parasitized larvae generally contained a single parasite but multiple infections also occurred. Upon emerging from the host (usually through the siphon), the nematodes entered the organic matter in the bottom of the treehole where they molted, mated, and oviposited. Under laboratory conditions, the final double molt occurred from 10–18 days after emergence with mating and oviposition occurring about 10–15 days after the final molt. The juveniles molted once within the egg and hatched about 15 days after oviposition.

Mermithids in various stages of their free-living development could be found throughout the year in the moist organic matter in the bottom of the treehole. By July, most of the eggs had been deposited and remained dormant over the fall and winter until the rains and warmer spring temperatures stimulated hatching.

Penetration of preparasitic juveniles into

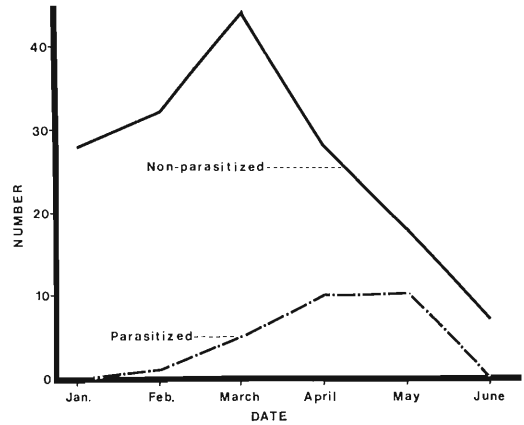


Figure 11. The number of mosquito larvae collected and parasitized by *Octomyomermis troglodytis* sp. n. during the winter and spring of 1972.

mosquito larvae was observed in the laboratory. The extremely active preparasitic juveniles, which spend most of their time at the surface of the water, made contact with resting *A. sierrensis* larvae. One parasite penetrated in the saddle area. Remaining motionless except for movement of its stylet, the mermithid pressed its head against the host's cuticle and pharyngeal gland secretions probably were expelled through the stylet onto the insect's cuticle. In about 10 min, a small hole was made in the mosquito's cuticle and the nematode suddenly straightened out and entered the hemocoel within a few seconds. Examination of newly penetrated juveniles showed that their penetration glands had collapsed, indicating their probable use for entry into the host.

The nematodes initiated growth in the host at the base of the siphon and then moved anteriorly after 8–10 days. The entire developmental period lasted from 20–22 days at 20 C. All hosts died soon after the nematodes emerged and detailed examinations indicated fat body depletion.

The number of parasitized larvae collected during the winter and spring of 1972 is shown in Figure 11. Most infected larvae were found in late March, April, and early May, when the mosquito populations were beginning to decline. In May, the incidence of parasitism reached 38%.

Discussion

Other pathogens of *A. sierrensis* in treeholes included the fungus, *Beauveria tenella*, and the ciliated protozoan, *Tetrahymena* sp. The latter organism was occasionally found developing together with *O. troglodytis* (Sanders, 1972).

A second mermithid species was recovered from sciarid larvae living in the bottom of the treehole during the summer months. This parasite was distinct from *O. troglodytis* and was never recovered from *A. sierrensis.*

Since *A. sierrensis* is often a problem in urban areas near gardens and parks, *O. troglodytis* may be considered a possible means of control for this and perhaps other treehole culicids. Although the natural distribution of this mermithid is apparently limited, techniques have been developed for the artificial propagation of mermithids of mosquitoes (Muspratt, 1965; Petersen and Willis, 1972) and it may be possible to mass produce *O. troglodytis* for release in uninfested treeholes. The mermithid, *Reesimermis nielseni* Tsai and Grundman, that is now being used for the biological control of mosquitoes, will not tolerate the water in treeholes and cannot be used against these species (Chapman, pers. comm.).

In the descriptive portion of this study, emphasis was placed on the morphology of the preparasitic (= infective) juveniles. In most mermithid descriptions, little, if any, mention is made of this stage. However, extensive examinations of the preparasitic juveniles of different mermithid species representing several genera showed that this stage contains characteristic taxonomic features. The length and shape of the stylet, shape of the penetration glands, length of the stichosome and intestine, position of the gonad primordium, presence of the anus, and length of the tail are basic diagnostic characters that can definitely be used in mermithid taxonomy.

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