Psammomermis nitidulensis n. sp. (Nematoda : Mermithidae) from sap beetles (Coleoptera : Nitidulidae) with biological observations and a key to the species of Psammomermis(1)

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Summary - Psammomermis nitidulensis n. sp. (Nematoda: Mermithidae) is described from three species of sap beetles, Carpophilus lugubris Murray, C. antiquus Melsh and Coloperta truncatus (Randall) (Coleoptera: Nitidulidae) from Mason County, IL, USA. This mermithid differs from previously described members of the genus by the shape of the amphids and their association with the amphidial glands, the length, form and curvature of the spicules and the shape of the vagina vera and vagina uterina. A key to the species of Psammomermis is provided and the generic diagnosis is emended. This mermithid causes up to 80 % parasitism and death of the dusky sap beetle (C. lugubris), an important vector of mycotoxigenic fungi. Investigations are underway to determine the possibility of utilizing this mermithid for biological control of this insect pest.

Resume - Psammomermis nitidulensis n. sp. (Nematoda : Mermithidae) provenant de trois coleoptères suceurs de sève (Coleoptera : Nitidulidae); observations biologiques et clé des espèces du genre Psammomermis - Psammomermis nitidulensis n. sp. (Nematoda : Mermithidae) est décrit provenant de trois espèces de coleoptères suceurs de sève, Carpophilus lugubris Murray, C. antiquus Melsh et Coloperta truncatus (Randall) (Coleoptera : Nitidulidae), récoltées dans le Comté de Mason, Illinois, États-Unis d'Amérique. Ce Mermithide diffère des autres espèces du genre par la forme des amphides et leur type d'association avec les glandes amphidiales, la longueur et la forme des vagina vera et vagina uterina. Une clé des espèces du genre Psammomermis est proposée et la diagnose en est amendée. Le Mermithide peut provoquer jusqu'à 80 % d'infestation et de mort chez le coleoptère suceur de sève C. lugubris, un important vecteur de champignons mycotoxigènes. Des recherches sont en cours concernant l'utilisation possible de ce Mermithide en vue du contrôle biologique de cet insecte nuisible.

Key-words : Carpophilus, dusky sap beetle, Nitidulidae, Psammomermis, nematodes.

Sap beetles (Coleoptera : Nitidulidae) are mostly small beetles less than 12 mm in length. Members of the family are attracted to fresh, processed and stored fruits and vegetables and some are pests. One member of this family, the dusky sap beetle, Carpophilus lugubris Murray, can damage a variety of vegetables, especially corn, where it occurs inside the kernels at the top of the ears. This species is also a significant vector of mycotoxigenic fungi and is known to carry the aflatoxin-producing fungus, Aspergillus flavus Link ex Fries to corn (Lussenhop & Wicklow, 1990).

A survey of various localities in Illinois, USA was initiated to search for natural enemies of C. lugubris and at one of nineteen sites surveyed over a 4-year period, a nematode was found parasitizing up to 80 % of the adults of the dusky sap beetle (Dowd et al., 1995).

The nematode proved to be an undescribed species of Psammomermis Polozhentsev, 1941 and is described below and biological observations and a key to the known species of Psammomermis are provided.

Materials and methods

Sap beetles were collected from the University of Illinois River Sand Field Experiment Station, Kilbourne, IL, USA from 1991 to 1994. The collection area was planted in corn and soybeans and was surrounded on two sides by oak forest and on the third side by open prairie. The individual insects were collected in PVC pipe traps (Dowd et al., 1992) baited with the pheromone of C. lugubris. The traps were examined once or twice a week and any adult sap beetles were removed from the traps and held on pinto bean-based diet in small plastic cups at room temperature and ambient light (Dowd, 1987). When the insects died, they were placed individually in small plastic vials containing 1 %

(1) Names are necessary to report factually on available data; however, the USDA neither guarantees nor warrants the standard of the product, and the use of the name by USDA implies no approval of the product to the exclusion of others that may also be suitable.
water agar and held until the nematodes emerged. The dead insects were removed after the nematodes had completely emerged and the nematodes were maintained in the agar at room temperature at ambient light and left until they had molted to the adult state. Adult nematodes were heat killed in water \(60^\circ\text{C}\), fixed in TAF and processed to glycerin.

**Psammomermis** Polozhentsev, 1941

**Diagnosis** (Kaiser, 1984; Poinar & Jackson, 1992; emended)

Long, thin terrestrial mermithids ranging in color from white to yellow; postparasitic and adult cuticle usually relatively thin, with faint or no cross-fibers under the light microscope at 20 x; head rounded to truncate, mouth opening terminal or shifted ventrad; lip papillae absent; six cephalic papillae arranged in one or rarely two planes; two hypodermal pegs may be present at the position of the tip of the head (these may be additional cephalic papillae or another type of sensory structure); amphids small to medium in size, from cup to pear to flask-shaped; opening circular to elliptical; eight hypodermal cords at midbody; spicules paired, separate, from medium to long (longer than body width at cloaca); vagina barrow or pear-shaped, muscular, often composed of an outer *vagina vera* portion and an inner *uterina* portion; vaginal canal straight or bent at the junction of *vagina vera* and *vagina uterina*; tail of postparasitic juvenile either entire, with a cuticular appendage or a small knob.

**Type species**

*P. korsakovi* Polozhentsev, 1941.

**Other species**

*P. acricephala* Ipatjeva & Pimenova, 1985  
*P. alechini* Artyukhovsky & Kharchenko, 1965  
*P. busuluk* Polozhentsev, 1952  
*P. byssina* Rubstov, 1976  
*P. canterburiensis* Poinar & Jackson, 1992  
*P. communis* Popov, 1978  
*P. conjuncta* Kaiser, 1984  
*P. cornicularis* Kaiser, 1984  
*P. filiformis* Kaiser, 1984  
*P. guffeldi* Ipatjeva & Pimenova, 1985  
*P. ipatjevae* Popov, 1978  
*P. kulaapi* Polozhentsev, 1941  
*P. montana* Ipatjeva & Pimenova, 1985  
*P. minor* Kaiser, 1984  
*P. nitidulensis* n. sp.  
*P. oesophaga* Artyukhovsky & Lisikova, 1977  
*P. parvula* Rubstov, 1976  
*P. pologenzevi* Popov, 1978  
*P. servicestidis* Baker, Poinar & Campbell, 1997

**Psammomermis nitidulensis** n. sp.  
(Fig. 1)

**Measurements**

- **Female** \((n = 7)\):  
  - \(L = 54 \ (26-85) \ mm\);  
  - greatest diameter = 104 \( (92-113) \ \mu m\);  
  - distance from head to nerve ring = 262 \( (238-276) \ \mu m\);  
  - length of amphidial pouch = 18 \( (16-19) \ \mu m\);  
  - diameter of amphidial opening = 3.8 \( (3.0-4.6) \ \mu m\);  
  - *vagina vera* length = 28 \( (19-32) \ \mu m\);  
  - *vagina uterina* length = 38 \( (29-45) \ \mu m\);  
  - \(V = 52 \ (49-56)\);  
  - distance from end of trophosome to tail tip = 140 \( (100-195) \ \mu m\).

- **Male** \((n = 6)\):  
  - \(L = 28 \ (20-42) \ mm\);  
  - greatest diameter = 78 \( (63-113) \ \mu m\);  
  - distance from head to nerve ring = 179 \( (174-190) \ mm\);  
  - length of amphidial pouch = 19 \( (15-22) \ \mu m\);  
  - diameter of amphidial opening = 3.2 \( (3.0-3.4) \ \mu m\);  
  - length of spicules = 261 \( (200-293) \ \mu m\);  
  - greatest width of spicule shaft = 5.6 \( (4.7-7.3) \ \mu m\);  
  - tail length = 170 \( (145-181) \ \mu m\);  
  - tail width at cloaca = 67 \( (60-73) \ \mu m\);  
  - ratio of tail length to tail width at cloaca = 2.6 \( (2.3-2.8)\).

*Fig. 1. Psammomermis nitidulensis* n. sp.  
*A*: Ventral view of male tail;  
*B*: Dorsal view of male head;  
*C*: Dorsal view of female head;  
*D*: Lateral view of female head;  
*E*: En face view of male;  
*F*: Lateral view of female tail;  
*G*: Cross section of male at mid-body;  
*H*: Lateral view of vulvar area;  
*I*: Lateral view of male tail with cuticles from last molts present (Scale bars: \(A, F = 32 \ \mu m\);  
\(B-D = 23 \ \mu m\);  
\(E = 13 \ \mu m\);  
\(G = 19 \ \mu m\);  
\(H-I = 38 \ \mu m\)).
DESCRIPTION

**Adults:** Narrow, small, white nematodes with a smooth cuticle. Flat to rounded head. Terminal mouth opening: six cephalic papillae arranged in a single circle, lacking distinct hypodermal cephalic pegs. Amphidial pouches flask to pear-shaped, closely associated with amphidial glands; amphidial openings circular, similar in both sexes; amphids located either slightly posterior or adjacent to the lateral cephalic papillae; eight hypodermal cords at mid-body; vagina *vra* (outermost portion of vagina adjacent to vulva) thin, lined with cuticle, meeting the vagina *uterina* (inner portion of vagina between vagina *vra* and the uterus) at a sharp angle; vagina *uterina* surrounded with strong circular muscles; vulva opening at a sharp angle. Spicules long, paired, separate, with a strong curve in the distal half, yellow, closely appressed in the distal half and tapering to a fine point; genital papillae in three rows; eight to eleven postcloacal papillae and fifteen to seventeen pre-cloacal papillae per row; pre-cloacal papillae extending past base of spicules; tail of both sexes narrowly rounded; small hypodermal peg transversing the cuticle of the tail in both sexes; disc-shaped protein crystals occurring in the body cavity of both sexes.

**Postparasitic juveniles (n = 10):** Cephalic papillae and amphids smaller and less conspicuous than in the adults; tail tip with a large cuticular projection, 40-55 μm in length.

An atypical condition occurred where the final molt consisted of not two but three separate cuticles (Fig. 11). It is not known whether one of the inner cuticles represented an extra molt or whether the cuticles represented the true second, third and fourth molts of the specimen. If the latter, then either the egg molt or the parasitic molt that is typical of mermithids, was surprised.

No cross fibers were noted in any of the cast cuticles.

**Type Host and Locality**

Dusky sap beetle, *Carpophilus lugubris* Murray (Coleoptera: Nitidulidae), collected from the Illinois River Sand Field Experiment Station at Kilbourne, Mason County, IL, USA. Other hosts were the smaller antique sap beetle (*Carpophilus antiquus* Melsh) and *Colopterus truncatus* (Randall), all of which are in the subfamily Carpophilinae of the Nitidulidae.

**Type Specimens**

Holotype (male) (UCDNC # 3319) and allotype (female) (UCDNC # 3320) deposited in the Division of Nematology, University of California, Davis, California. Paratypes in the collection of the Muséum National d'Histoire Naturelle, Paris, and in the senior author's collection.

**Diagnosis and Relationships**

Small, narrow nematode with six cephalic papillae, eight hypodermal cords and medium-sized amphids. Both the adult and the postparasitic juvenile cuticles lack cross-fibers. Key characters separating the present species from previously described ones are the ratio and angle of the vagina *vra* to the vagina *uterina*, the amphidial glands in association with the amphidial pouches, the length and shape of the spicules, the length/width ratio of the male tail—a character used by Artyukhovski (1990) for separating members of this genus—the area covered by the genital papillae (especially pre-cloacal), the terminally placed mouth and the absence of cephalic hypodermal pegs.

**Biological Observations and Discussion**

Most of the postparasites of *P. nitidulensis* emerged from their adult beetle hosts within 2 weeks after field collections. The final molt to the adult stage occurred between 2 and 3 months later. Normally there was only a single nematode per host but as many as four nematodes were observed to emerge from a single beetle.

Three separate beetle species can serve as hosts to *P. nitidulensis* n. sp. However almost all of the nematodes emerging from *C. lugubris* were female while nematodes emerging from *C. antiquus* and *C. truncatus* were both male and female. Environmental control of sex in the Mermithidae is well known (Poinar & Hansen, 1984) and it is generally considered that an abundance of nourishment results in female production whereas restricted nourishment (as a result of small hosts or multiple infections in a single host) results in male formation. In the case of *Hexamermis arvalis* Poinar & Gyrisco, 1962, infection of alfalfa weevil larvae resulted in the production of almost all males (Poinar & Gyrisco, 1962). This could have been an indication of a relatively recent host-parasite association. The alfalfa weevil (*Hypera postica* Gyll.) was a recent European introduction into the area where *H. arvalis* normally parasitized plant bugs and noctuid larvae. Parasitizing the alfalfa weevil without a period of adaptation resulted in the formation of males, perhaps because some nutritional unbalance affected female development. The same scenario may occur in the present situation. Just when *C. lugubris* entered North America is not known but since it is primarily a southern and neotropical species (Blackwell, 1945), introductions could have been less than 100 years ago. In this case, *C. lugubris* would be a recent host without any period of adaptation, resulting in the formation of predominately females. In contrast, the nematode had already established a balance with the smaller native species, *C. antiquus* in which it produced both sexes. It appears that mermithids, as with entomogenous nematode parasites in general, are able to adapt quickly to new hosts in the environment. However, several stages of adaptation are required, including an adjustment to the physiological conditions in the host's hemocoel, avoiding host reactions, completing parasite development and establishing a normal sex ratio. In the present case, parasitism of *C. lugubris* is made possible since the
native host species can provide males. As long as at least one other of the three hosts is present with C. lugubris, a normal parasitic cycle should continue. After a period of time, a physiological race of P. nitidulensis n. sp. would be expected to arise which would produce both sexes in C. lugubris.

Key to the species of Psammomermis

The following key separates the present species from previously described members of Psammomermis. Diagnostic characters of many of the Russian species were obtained from Aryanovskii (1990). Since basic diagnostic characters (including males) are absent from the description of Popov 1978, it is omitted from the key.

1 - Head with one or two hypodermal pegs in addition to six cephalic papilae ................................................. 2
   2 - Head without hypodermal pegs .................................. 5

2 - Head with a single dorsal hypodermal peg ..................... 3
   3 - Head with a pair of hypodermal pegs ......................... 4

3 - Female length over 100 mm; female head flat
   - Female length under 50 mm; female head attenuated .........
   ........................................................................... P. sericechadis

4 - Hypodermal pegs in lateral position .............................. P. corniculalis
   - Hypodermal pegs in dorsal position ............................... P. communis

5 - Spicules short, under 200 J.Lm in length ....................... 6
   6 - Spicules greater than 200 J.Lm in length ..................... 11

6 - Spicule length less than 75 J.Lm ................................. P. guffeldi
   7 - Spicule length 75-200 J.Lm ................................. 7

7 - Mouth ventral .......................................................... P. parva
   - Mouth terminal ......................................................... 8

8 - Vagina vera meeting vagina uterina at an angle ................. P. montana
   - Vagina vera and vagina uterina form a straight or nearly a straight line ................................................... 9

9 - Vagina vera longer than vagina uterina .......................... P. cantbruirensis
   - Vagina vera shorter than vagina uterina ....................... 10

10 - Spicule length 75-100 J.Lm; adult head not pointed .........
    ........................................................................... P. bysiina
    - Spicule length 100-200 J.Lm; adult head pointed ..........
    ........................................................................... P. minor

11 - Spicule length 200-450 J.Lm ....................................... 12
    - Spicule length > 450 J.Lm ......................................... 17

12 - Ratio of tail length over tail width 1.5-3.0 ...................... 13
    - Ratio of tail length over tail width 0.5-1.5 ........................ 13

13 - Ratio of tail length over tail width 2.3-3.0 ...................... 14
    - Ratio of tail length over tail width 1.5-2.3 ...................... 15

14 - Vagina vera meeting vagina uterina at an angle; spicule length < 300 J.Lm ............................................. P. nitidulensis n.sp.
    - Vagina vera forming a straight line with vagina uterina; spicule length > 300 J.Lm ........................................... P. alechini

15 - Spicule length 200-300 J.Lm ....................................... P. kilagini
    - Spicule length 300-450 J.Lm ....................................... 16

16 - Vagina vera meeting vagina uterina at an angle; spicule curved twice ..................................................... P. conjuncta
    - Vagina vera forming a straight line with vagina uterina; spicule curved once ............................................. P. korsakovi

17 - Ratio of tail length over tail width = 1.0 ...................... P. busuluk
    - Ratio of tail length over tail width > 1.5 ......................... 18

18 - Spicules curved twice ............................................. P. pologenzevi
    - Spicules with single curve ......................................... P. filiformis

Discussion

The genus Psammomermis is cosmopolitan in distribution, having been recorded from several land masses including New Zealand and Australia. Known hosts are limited to members of the Coleoptera, especially the Scarabaeidae. This is the first report of an infection with members of the family Nitetulidae. The genus Psammomermis was erected by Polozhentsev (1941) when he discovered terrestrial mermithids that fit in the aquatic genus Mesomermis. Thus, the original diagnostic character separating the two genera was that Psammomermis was terrestrial. This ecological distinction met with criticism amongst other workers and Welch (1956) did not recognize the genus since he felt that Polozhentsev was following the teachings of Lysenko and Michurin by erecting Psammomermis. As a result Psammomermis was and sometimes still is synonymized with Mesomermis (Poinar, 1977).

However, a re-evaluation of the characters in the growing number of species showed that members of the genus Psammomermis share common characters and that the narrow body shape, form of the amphids, and shape and size of the spicules, differ from the species placed in Mesomermis. This is not to say that variation does not exist among and between species of Psammomermis. The presence of cephalic hypodermal pegs (function unknown) in some of the species in the genus is one example. As with other mermithid genera, adaptation to new hosts is an important aspect of their evolution and just how different hosts affect character formation in mermithids is a relatively unexplored area.

Some species of Psammomermis have been described as possessing an “excretory pore” near the nerve ring. The present species lacks such a pore. In general, mermithids usually lack excretory pores, yet structures resembling excretory pores have been described in species from several genera. Whether this structure is really associated with an excretory system still awaits proof. There is some evidence suggesting that these “excretory pores” are in fact nerve endings that penetrate the cuticle (Poinar, unpubl.). Further studies on these structures are desirable.

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References


