Description and SEM Observations of *Meloidogyne sasseri* n. sp. (Nematoda: Meloidogynidae), Parasitizing Beachgrasses

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Abstract: *Meloidogyne sasseri* n. sp. is described and illustrated from American beachgrass (*Ammophila breviligulata*) originally collected from Henlopen State Park and Fenwick Island near the Maryland state line in Delaware, United States (6). Its relationship to *M. graminis*, *M. spartinae*, and *M. californiensis* is discussed. Primary distinctive characters of the female perineal pattern were a high to rounded arch with shoulders, widely spaced lateral lines interrupting transverse striations, a sunken vulva and anus, and coarse broken striae around the anal area. Second-stage juvenile body length was 554 μm (470–650), stylet length 14 μm (13–14.5), tail length 93 μm (83–115), tapering to a finely rounded terminus. Male stylet length 20 μm (19–21.5), spicule length 33 μm (30–36). Scanning electron microscope observations provided additional details of perineal patterns and face views of the female, male, and J2 head. Wheat, rice, oat, *Ammophila* sp., *Panicum* sp., bermudagrass, zoysiagrass and St. Augustinegrass were tested as hosts. Distribution of the species was the coasts of Delaware and Maryland. The common name “beachgrass root-knot” is proposed for *M. sasseri* n. sp.


In August 1990, a root-knot nematode was found parasitizing the roots of beachgrasses, *Ammophila breviligulata* Fern. and *Panicum amarulum* Hitchcock & Chase, at Henlopen State Park and Fenwick Island near the Maryland state line in Delaware (6). The nematode did not produce galls, and females were generally surrounded by a massive egg sac. Morphological studies proved that the species discovered in Delaware is different from others in the genus *Meloidogyne*; it is described herein as *M. sasseri* n. sp. Additionally, we present results of limited host-range tests on a few economically important grass species and also give the distribution of this new species.

**Materials and Methods**

Cultures on beachgrasses (*Ammophila breviligulata* and *Panicum amarulum*) were originally collected in August 1990 (6) at Henlopen State Park and Fenwick Island in Delaware, near the Maryland state line, and were maintained in the greenhouse as stock cultures on the same hosts at Beltsville. Nematode reproduction was evaluated on nine graminaceous plants in growth chambers at 25–27°C with 14 hour/10 hour light/darkness cycles: wheat (*Triticum aestivum* L. cvs. Coldwell and F1 302), rice (*Oryza sativa* L. cv. Lemont), oat (*Avena sativa* L. cv. Lemont), corn (*Zea mays* L.), beachgrass (*Panicum amarulum*), American beachgrass (*Ammophila breviligulata*), bermudagrass (*Cynodon dactylon* (L.) Pers.), zoysiagrass (*Zoysia japonica* Steudal), and St. Augustinegrass (*Stenotaphrum secundatum* (Walter) Kuntze). Plants were grown in steam-sterilized, 15-cm-d clay pots containing steam-sterilized Astatula fine sand and subsequently inoculated with 500 second-stage juveniles (J2). Inoculum consisted of J2 obtained from egg masses kept in petri dishes with a small amount of water. Twenty ml of aqueous suspension containing 500 J2 was pipeted into each pot in the soil around the roots of 1-week-old plants. The experiment was terminated 90 days after inoculation, and the test was repeated twice.

For morphological observation, J2 and males were recovered from fresh infected...
roots or egg sacs kept in petri dishes with a small amount of water. Some were recovered from soil by sieving and Baermann funnel. Females were dissected from infected roots after fixation overnight in 3% formaldehyde.

Procedures used in measuring and preparing specimens were essentially those used by Golden and Birchfield (3), except some fixed females were cut and mounted in a clear lactophenol solution. Photomicrographs of perineal patterns, J2, and males were made with an automatic 35-mm camera attached to a compound microscope having an interference contrast system. Roots and whole females were photographed with an automatic 35-mm camera attached to a dissecting microscope. For scanning electron microscopy (SEM), living specimens were fixed in 3% glutaraldehyde solution buffered with 0.05 M phosphate (pH 6.8), dehydrated in a graded series of ethanol, critical-point dried from liquid CO2, and sputter-coated with a 20-30-nm layer of gold-palladium. All measurements are in micrometers (µm) unless otherwise stated. When ranges are given, mean values followed by the standard deviation (SD) are often given within parentheses.

**Systematics**

*Meloidogyne sasseri* n. sp.

(Figs. 1–8)

**Description**

*Holotype (female, in glycerine):* Body length with neck 822; body width 493; neck length 110, neck greatest width 46; stylet 14; stylet knob width 5; stylet knob height 2; dorsal esophageal gland orifice (DGO) from base of stylet 4; excretory pore from anterior end 13.5; EP/ST ratio 1.0; body length from anterior end to posterior end of metacorpus 172; about 10 body annules from anterior end to excretory pore; cuticle thickness at neck 8; cuticle thickness at midbody 30; vulval slit length 30; distance from vulval slit to anus 18.

*Female (n = 60):* Body length including neck 644–1,150 (860; SD 120.7); neck length 60–140 (106; SD 18.7); body width 395–685 (524.5; SD 72.6); neck width 30–70 (43.5; SD 7.4); cuticle thickness at neck 7–14 (9.9; SD 1.7); cuticle thickness at midbody 16–42 (26.5; SD 5.6); a = 1.3–2.4 (1.6; SD 0.2); stylet length 13–15 (14; SD 0.6); style knobs width 4–5.5 (4.8; SD 0.4); stylet knobs height 1.5–2.5 (2.0; SD 0.2); DGO from base of stylet 3.0–5.5 (4.2; SD 0.6); excretory pore from anterior end 7.5–31 (15.2; SD 2.9); EP/ST ratio 0.5–2.2 (1.3; SD 0.3); body length from anterior end to posterior end of metacorpus 106–194 (148.1; SD 20.3); excretory pore about 7–15 (11.6; SD 1.8) annules from anterior end; vulval slit length 25–35 (30.2; SD 2.3); distance from vulval slit to anus 13–22 (16.9; SD 2.4).

Body white, round to pear shaped, with slight posterior protuberance; neck long (Fig. 2C), distinct, tapering anteriorly. Cephalic framework weak, slightly offset from neck, bearing one large smooth annule. Lip region variable in shape with circumoral elevation. Labial disc slightly raised above medial lips (Fig. 2B), disc and medial lips dumbbell-shaped. Lateral lips smaller than and adjacent to medial lips. Amphidial openings oval, located between labial disc and lateral lips. Stylet strong, basal knobs heavy, rounded, sloping posteriorly (Fig. 6B,C). Excretory pore distinct, generally located posterior to stylet, sometimes at level of or above unprotruded stylet base. Esophagus well developed; procorpus long, cylindrical and metacorpus spherical with heavy sclerotized valve. Cuticle thick at neck and midbody. Perineal pattern (Figs. 1A–F, 5A–I) usually with high to rounded arch with shoulders and widely spaced lateral line interrupting transverse striations; vulva and anus sunken with coarse broken striae around and near the vulval area. Phasmids small, nor prominent, seen only in some specimens (Fig. 5I). Eggs deposited in a gelatinous matrix.

*Allotype (male in glycerine):* Length = 1,700; a = 37; b = 9.1; c = 213; stylet length 20; stylet knobs width 5; stylet
knobs height 3; excretory pore from anterior end 140; center of median bulb 94 from anterior end; spicules 32; gubernaculum 8.5; tail 8.

**Male (n = 45):** Length 1,290–2,175 (1,746; SD 192.6); width 30–50 (43.8; SD 4.1); a = 32.2–51.7 (39.9; SD 3.6); b = 6.1–12.9 (8.6; SD 13); c = 161–257 (203.2;...
Fig. 2. SEM micrographs of females and eggs of *Meloidogyne sasseri* n. sp. A) Anterior region showing excretory pore (arrow). B) Face view (arrow = excretory pore). C) Whole female. D) Eggs.
Fig. 4. SEM micrographs of second-stage juveniles of *Meloidogyne sasserii* n. sp. A) Lateral view of head region. B,C) Enface view. D) Posterior region. E,F) Lateral field.
Fig. 5. Photomicrographs of perineal patterns of Meloidogyne sasseri n. sp. I) Showing phasmids (arrows).
Fig. 7. Photomicrographs of second-stage juveniles and eggs of Meloidogyne sasseri n. sp. A,B) Anterior regions. C) Part of anterior region showing excretory pore (arrow). D) Basal esophageal bulb showing three nuclei (arrows). E) Lateral field. F) Eggs. G–J) Posterior regions showing inflated rectum (arrows).
**Fig. 8.** Photomicrographs of specimens of *Meloidogyne sasseri* n. sp. on or isolated from the following roots: A,B) Bermudagrass. C) Panicum. D) Zoysiagrass. E) Rice. F) Wheat. G) Females dissected from roots. H) Females with egg mass attached.
Body slender, vermiform, tapering gradually towards ends. Head slightly offset, labial disc in face view slightly rounded, raised above crescentic medial lips extending some distance into head region. Each medial lip has one pair of faint cephalic sensilla, marked by small cuticular depressions. Amphidial openings appear as long slits. Cuticular annulation distinct. Annules 2.5–3.5 wide at midbody, becoming smaller towards ends of body. Midbody width averaging 44. Lateral field with four incisures, aerolated (Fig. 3B). Stylet heavy, knobs massive, rounded, sloping posteriorly. Hemizonid prominent, about two annules long, one annule posterior to excretory pore. Excretory pore usually near middle of basal esophageal bulb, in some specimens more posteriorly. Phasmids about 6 from tail terminus. Spicules arcuate, tips rounded (Fig. 3E), gubernaculum short, simple. Tail rounded.

Second-stage juvenile: Measurements of 350 juveniles in Table 1.

Body cylindrical, vermiform, tapering at both extremities, more so posteriorly (Fig. 7G–J). Head truncate, slightly set off, without annulation; cephalic framework weak. Labial disc slightly raised above medial lips, lateral lips in same contour with head region. Labial disc and medial lips fused, dumbbell shaped in face view. Stylet large, heavy, knobs rounded, usually sloping posteriorly. Body annulation fine, distinct. Lateral field with four incisures, aerolated (Fig. 4E,F). Excretory pore below nerve ring in isthmus region. Hemizonid located 1–2 annules posterior to excretory pore. Rectum inflated (Fig. 7H,J). Tail very long, tapering gradually to finely rounded or variable shaped terminus.

Egg (n = 40): Length 88–136 (113.1; SD 9.9); width 44–60 (49.3; SD 3.8); L/W ratio 1.7–2.9 (2.2; SD 0.2); egg shell hyaline, without markings.

Type host and locality

Roots of American beachgrass (Ammophila breviligulata Fern.) from Henlopen State Park in Delaware, USA.

Type specimens

Holotype (female): Isolated from growth chamber culture, propagated on American beachgrass (Ammophila breviligulata), derived from original population from the above type locality. Slide T-497t, deposited in the U.S. Department of Agriculture Nematode Collection, Beltsville, Maryland, USA.

Allotype (male): Slide T-498t, same data and repository as holotype.

Paratypes (females, males, and juveniles): Same data and repository as holotype. Others deposited in University of California Nematode Collection (UCRNC), Riverside, California; The Nematode Collection of the Nematology Department, Rothamsted Experimental Station, Harpenden, Herts., England; Canadian National Collection of Nematodes; Ottawa, Canada; Collection Nationale de Nematodes, Laboratoire des Vers, Muséum national d’Histoire naturelle, Paris, France; Nematode Collection, Institutut voor Dierkunde, Laboratorium voor Morfologie en Systematiek der Dieren, Gent, Belgium; Nematode Collection of the Landbouwhogeschool, Wageningen, The Netherlands; Commonwealth Institute of Parasitology Collection, St. Albans, Herts., England.

Diagnosis

Meloidogyne sasseri n. sp. is characterized by having a female perineal pattern with a high to rounded arch with occasional shoulders, and widely spaced lateral line interrupting transverse striae, sunken vulva, and anus and coarse broken striae around and near the anal area. J2 have a
### Table 1. Measurements (μm) of second-stage juveniles of *Meloidogyne sasseri* n. sp. from seven hosts.

<table>
<thead>
<tr>
<th>Characters</th>
<th>Ammophila (n = 50)</th>
<th>Panicozum (n = 50)</th>
<th>Rice (n = 50)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Body length</td>
<td>490.0-575.0</td>
<td>470.0-640.0</td>
<td>485.0-610.0</td>
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<tr>
<td>Body width at midbody</td>
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<td>16.0-20.0</td>
<td>16.0-20.0</td>
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<td>Head width (HW)</td>
<td>5.0-6.5</td>
<td>5.0-6.0</td>
<td>4.5-6.0</td>
</tr>
<tr>
<td>Head height (HH)</td>
<td>2.0-3.0</td>
<td>2.5-3.0</td>
<td>2.5-3.0</td>
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<tr>
<td>HW/HH ratio</td>
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<td>1.8-2.4</td>
<td>1.6-2.4</td>
</tr>
<tr>
<td>a</td>
<td>25.0-32.8</td>
<td>27.2-35.9</td>
<td>28.3-35.8</td>
</tr>
<tr>
<td>b</td>
<td>2.0-3.0</td>
<td>2.2-3.3</td>
<td>2.0-2.6</td>
</tr>
<tr>
<td>c</td>
<td>5.2-6.3</td>
<td>5.0-6.5</td>
<td>5.2-6.3</td>
</tr>
<tr>
<td>Stylet length</td>
<td>10.0-14.5</td>
<td>13.0-14.5</td>
<td>13.0-14.5</td>
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<tr>
<td>Base of stylet to DGO</td>
<td>1.5-3.0</td>
<td>2.5-4.0</td>
<td>2.5-3.5</td>
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<tr>
<td>Head tip to median bulb valve</td>
<td>60.0-68.0</td>
<td>55.0-80.0</td>
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<td>Head tip to base of esophageal gland lobe</td>
<td>175.0-257.0</td>
<td>170.0-265.0</td>
<td>204.0-250.0</td>
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<tr>
<td>Tail length</td>
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<td>85.0-106.0</td>
<td>85.0-104.0</td>
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<td>Hyaline tail terminal</td>
<td>16.0-23.0</td>
<td>16.5-27.0</td>
<td>15.0-24.0</td>
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<td>Caudal ratio A</td>
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<td>3.0-5.5</td>
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<tr>
<td>Caudal ratio B</td>
<td>5.7-10.0</td>
<td>7.2-14.7</td>
<td>5.6-11.5</td>
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<td>Width of tail at hyaline portion</td>
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<td>4.0-5.5</td>
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<tr>
<td>Width of tail 5 μm from tail tip</td>
<td>2.0-3.0</td>
<td>1.5-3.0</td>
<td>2.0-3.0</td>
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</tbody>
</table>

### Wheat (n = 50), Oat (n = 50), Bermudagrass (n = 50), Zoysiagrass (n = 50)
mean body length of 554 with a large, heavy stylet of about 14; long tail 93 (83–115), tapering to a finely rounded terminus. Male stylet averaging 20 with massive rounded knobs, spicule mean of 30; lateral field in juveniles, and males with four incisures.

Relationship

This new species is morphologically similar to *M. graminis* (Sledge & Golden, 1964) Whitehead, 1968 (7), *M. sartinae* Rau & Fassuliotis, 1965 (5), and *M. californiensis* Rahman & Maggenti, 1987 (1). From *M. graminis* it differs by having, in J2: longer body length 552 (470–650) vs. 475 (420–510); larger and heavier stylet 14 (13–14.5) vs. 12.6 (11.7–13.4); longer tail 96 (83–115) vs. 78 (68–88); in females: a sunken vulva and anus vs. situated on a slight but distinct button-like protrusion of the body; longer stylet 14 (13–14.5) vs. 12.6 (11.7–13.4); in males: a longer styler and spicule 20 (19–21.5), 33 (30–36) vs. 18.3 (17.9–19), 28.2 (28–29.1), respectively. Also, St. Augustinegrass is a poor host for *M. sartinae* n. sp. but is an excellent host for *M. graminis*, the type host. From *M. sartinae* it differs in having a J2 stylet mean of 14 vs. 15.4, shorter body length 552 (470–650) vs. 778 (612–912), location of hemizonid in relation to excretory pore 4 vs. 20 posterior to excretory pore, tail tapering to a finely rounded terminus vs. asymmetrical, spiked bulbous tail; in females, the perineal pattern has a high to round arch with occasional shoulders, widely spaced lateral line interrupting transverse striae, sunken vulva and anus and coarse broken striae around and near anal area; and hemizonid in male and J2 is posterior to excretory pore. *Meloidogyne californiensis* females have a shorter body length, cuticle at midbody only 3 thick, delicate stylet, prominent posterior cuticular protuberance, perineal pattern marked by one prominent stria in perineum, indistinct lateral lines and many broken discontinuous striae on both sides of the arch; and hemizonid in males and J2 is anterior to excretory pore.

DISCUSSION

Morphometrics of *Meloidogyne sartinae* n. sp. on *Panicum amarulum* and *Ammophila breviligulata* from Delaware, and from the populations obtained from cultures originating from these plants at Beltsville (Table 1), had both short- and long-tail forms in J2. Although initially we thought these forms represented two different species, our single egg mass cultures also had both long and short-tail forms, indicating conspecificity. Because the only difference was in tail length, we considered this to be normal variation within the species. This kind of variation has been observed in the *M. graminis* complex on bermudagrass and zoysiagrass (2).

Reexamination of specimens in the USDA Nematode Collection submitted by L. R. Krusberg (University of Maryland) and tentatively identified by Dr. Golden in 1970 as *M. graminis* on *A. breviligulata* from Ocean City, Maryland (with a note, "Larvae longer than normal") confirmed them to be *M. sartinae* n. sp. There are a few reports of an unknown root-knot nematode (*Meloidogyne* sp.) on American beachgrass along the coast of North Carolina (8,9). We believe the limited measurements of larvae of *Meloidogyne* sp. given by Young and Lucas (8) on *A. breviligulata* represent the present species, but this material no longer exists (L. D. Young, pers. comm.). Recently, Jepson (4) described *M. maritima* on
Ammophila arenaria in Perranporth, Cornwall, England.

In the host-range test, *M. sasser* reproduced on wheat, rice, oat, American beachgrass, bermudagrass, zoysiagrass, and St. Augustinegrass. The nematodes did not reproduce on corn. Some were better hosts than others. St. Augustinegrass was a poor host, although it is an excellent host for *M. graminis* (7). *Meloidogyne sasseri* n. sp. did not cause galling on the hosts in these tests, and their bodies protruded from the roots with a large egg mass attached at the posterior end (Fig. 8A–F,H). This response is similar to that of the *Meloidogyne* sp. reported by Young and Lucas (8) from North Carolina, further supporting our premise that this North Carolina *Meloidogyne* sp. is *M. sasser*. Interestingly, the morphometrics of J2 from the zoysiagrass population were relatively longer than those from the other seven graminaceous hosts. The J2 on oat and rice showed the shortest range of morphometric data. Also, because differences were not noted in the morphometrics of females and males from the other seven graminaceous populations, representative data from *Panicum amarulum* only was taken into account.

The species name is given in honor of Professor Dr. J. N. Sasser for his outstanding contributions to our knowledge of root-knot nematodes and also for his dedicated research and leadership as project leader of the International *Meloidogyne* Project on a worldwide basis.

**Literature Cited**


