

# **Noctuidonema dibolia** n. sp. (Aphelenchida: Acugutturidae), an Ectoparasite of the moth *Mocis latipes* (Lepidoptera: Noctuidae)<sup>1</sup>

O. G. MARTI, JR. AND C. E. ROGERS<sup>2</sup>

**Abstract:** *Noctuidonema dibolia* n. sp., an ectoparasite of adults of the noctuid moth *Mocis latipes* (Guenée) is described. The differentiating characters are a club-shaped body with a subterminal vulva in the female, spicules with a reduced matrix and sheath and closely apposed dorsal and ventral arms in the male, very long stylet and conus, moderately prominent stylet knobs, a bluntly rounded head, and a large renette cell in both sexes. Lateral fields, rectum, anus, bursa, and gubernaculum are absent. *Noctuidonema dibolia* differs from the other species of the subfamily Noctuidonematinae in the size and robustness of the body, the length of the stylet and conus, the length of the tail, and the shape of the spicules.

**Key words:** ectoparasite, insect parasite, Lepidoptera, *Mocis latipes*, moth, nematode, Noctuidae, *Noctuidonema dibolia* n. sp., taxonomy.

Surveys of Lepidoptera in French Guiana (9,12), the tropical Americas (11,12), and the southeastern United States (10) documented the widespread occurrence of an ectoparasitic nematode, *Noctuidonema guyanense* Remillet and Silvain, particularly among moths in the family Noctuidae. *Noctuidonema* Remillet and Silvain is of interest because it is one of only a few known metazoan parasites of adult moths (3) and is currently under study as a possible biological control agent for the fall armyworm moth, *Spodoptera frugiperda* (J. E. Smith), a serious pest of corn, sorghum, and pasture grasses (12,13).

*Mocis latipes* (Guenée), whose larvae are known as striped grass loopers, is primarily a pest of pasture grasses (5,12). Specimens of the genus *Noctuidonema*, collected from *M. latipes* in Tift County, Georgia, were examined and found to differ from *N. guyanense*, *Vampyronema daptria* (Anderson and Laumond) (2,7), and the related genus *Acugutturus* Hunt (6), and are here described as a new species, *N. dibolia*. The specific name 'dibolia' is taken from Greek (= pointed at both ends).

## MATERIALS AND METHODS

*Mocis latipes* adults were collected from 1991-93 in a walk-in ultraviolet light trap located on a research farm near Tifton, Georgia. The external surfaces of the moth abdomens were examined with a stereoscopic microscope for the presence of nematodes. Infested moths were placed into a 15-ml cup containing 2 ml of water plus a few drops of Tween 80 (polyoxyethylenesorbitan monooleate) and shaken for a few seconds to dislodge nematodes. Nematodes were concentrated by sedimentation in distilled water, examined live or killed in hot water (60 C), and fixed and stored in TAF (Triethanolamine-Formalin-Water 7:2:91) (4) until processed through an alcohol series to glycerin. The nematodes were then mounted on glass slides and examined with differential interference contrast or phase contrast optics. Drawings are based on microscopic examination and photographs of living as well as processed specimens. Values in the text are expressed as: range in  $\mu\text{m}$  (mean in  $\mu\text{m} \pm \text{SD}$ ).

## SYSTEMATICS

*Noctuidonema dibolia* n. sp.  
(Figs. 1A-C; 2A-E)

**Holotype (female):** Length = 522  $\mu\text{m}$ ; maximum width = 57  $\mu\text{m}$ ; width at vulva = 44  $\mu\text{m}$ ; anterior end to excretory pore

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<sup>1</sup> Mention of a trade name, warranty, proprietary product, or vendor does not constitute an endorsement of a product and does not imply its approval to the exclusion of other products or vendors that may also be suitable.

<sup>2</sup> Microbiologist and Research Entomologist, USDA, ARS, Insect Biology and Population Management Research Laboratory, P.O. Box 748, Georgia Coastal Plain Experiment Station, Tifton, GA 31793.

= 22  $\mu\text{m}$ ; anterior end to nerve ring = 137  $\mu\text{m}$ ; stylet length = 108  $\mu\text{m}$ ; stylet conus length = 91  $\mu\text{m}$ ; metacarpus length = 28  $\mu\text{m}$ ; metacarpus width = 23  $\mu\text{m}$ ; ovary length = 225  $\mu\text{m}$ ; tail length = 66  $\mu\text{m}$ .

*Allotype (male)*: Length = 464  $\mu\text{m}$ ; maximum width = 41  $\mu\text{m}$ ; width at cloaca = 12  $\mu\text{m}$ .

16  $\mu\text{m}$ ; anterior end to excretory pore = 25  $\mu\text{m}$ ; anterior end to nerve ring = 118  $\mu\text{m}$ ; stylet length = 89  $\mu\text{m}$ ; stylet conus length = 73  $\mu\text{m}$ ; metacarpus length = 28  $\mu\text{m}$ ; metacarpus width = 20  $\mu\text{m}$ ; testis length = 225  $\mu\text{m}$ ; tail length = 8  $\mu\text{m}$ ; spicule length = 44  $\mu\text{m}$ ; rostrum length = 12  $\mu\text{m}$ .

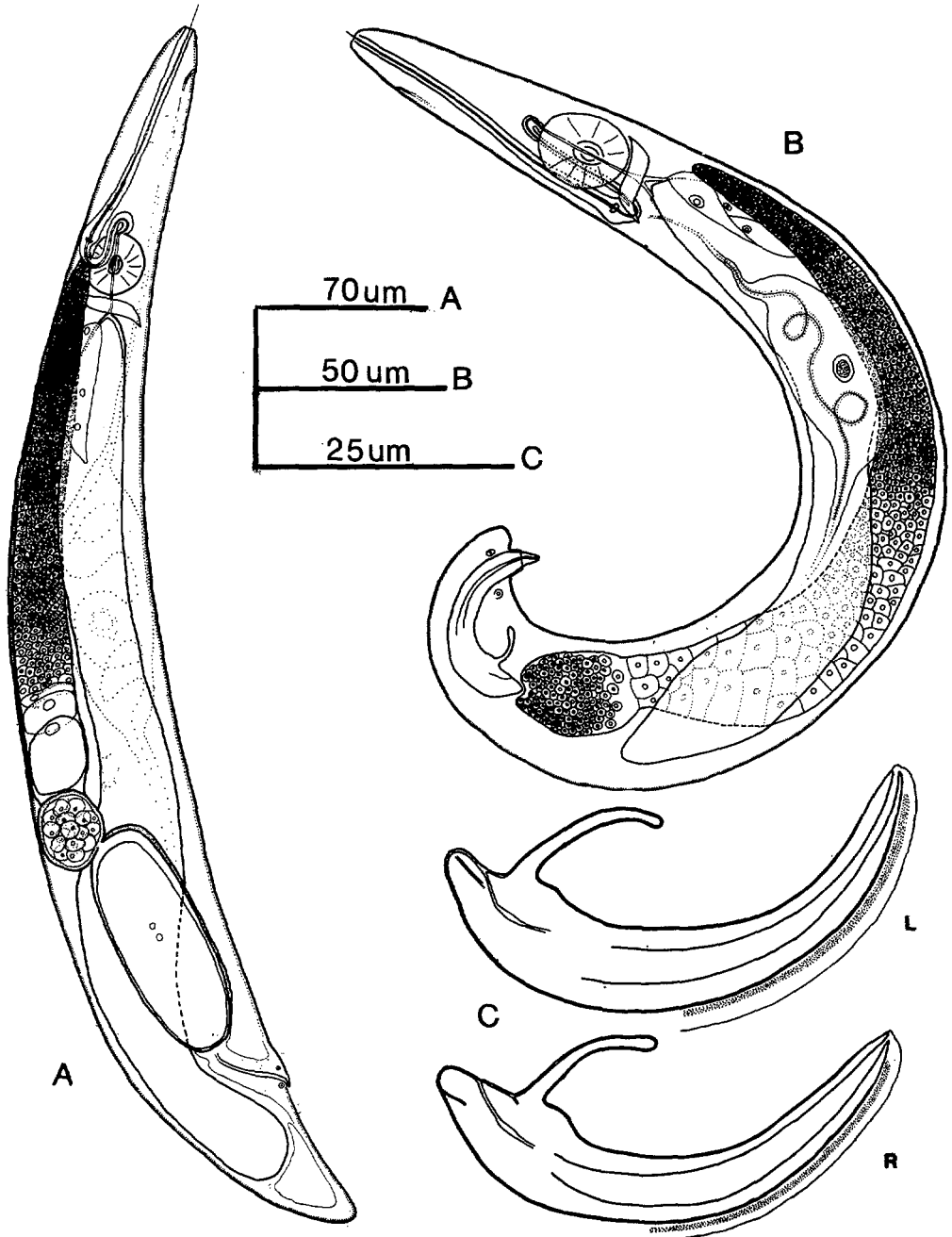


FIG. 1. *Noctuidonema dibolia* n. sp. A) Female. B) Male. C) Spicules.

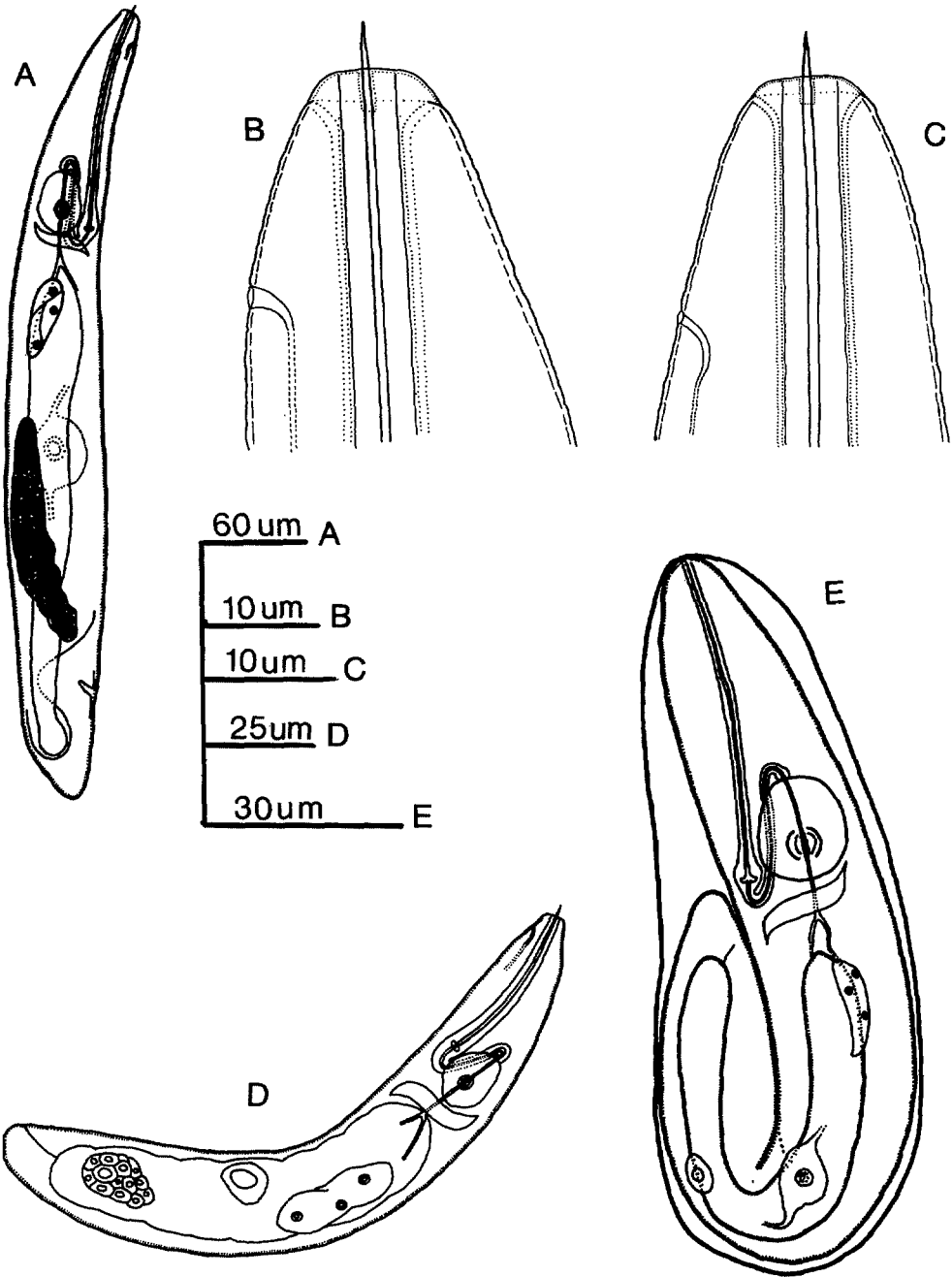


FIG. 2. *Noctuidonema dibolia* n. sp. A) Fourth-stage juvenile. B) Head end of female. C) Head end of male. D) Newly hatched second-stage juvenile. E) Embryonated egg with fully formed juvenile.

*Paratypes*: Measurements of 39 females and 16 males in Tables 1–4.

*Description*

*Female*: Measurements in Tables 1–3. Body club-like, slightly arcuate ventrally. Annules continuous, 1.0–1.6 wide ( $1.3 \pm$

0.2) at midbody, anastomoses common. Lateral fields absent. Lip region low, bluntly rounded, not strongly set off. Stylet flexible, delicate, elongate, representing 19–22% of total body length. Conus representing 80–82% of total stylet length. Conus midpoint diameter 0.8–1.0

TABLE 1. Measurements of 14 small paratype (<500  $\mu\text{m}$  body length), non-gravid female *Noctuidonema dibolia* n. sp. from Tift County, Georgia.

Character <sup>a</sup>	Mean	SD	Range
Measurements ( $\mu\text{m}$ )			
Body length	455	37.2	396–495
Maximum body width	54	7.3	44–67
Body width at nerve ring	36	3.1	32–41
Body width at vulva	41	5.9	34–54
Body width at anterior end of intestine	36	3.1	32–41
Anterior end to base of bulb	120	9.8	105–140
Metacarpus length	27	2.0	23–31
Metacarpus width	23	1.8	20–26
Excretory pore to anterior end	18	2.9	15–23
Stylet total length	99	6.1	83–108
Stylet conus length	81	5.3	69–89
Stylet shaft length	18	3.0	15–23
Head width	9	0.8	7–10
Head length	4	0.5	3–4
Ovary length	200	37.4	149–262
Vulva to end of tail	53	7.4	42–70
Vulva to anterior end	402	33.4	346–433
Vagina depth	24	4.1	18–31
Anterior end to anterior ovary	129	25.4	85–187
Ratios and Percentages			
a	8.5	0.7	7–10
b	3.8	0.4	3–5
c	8.6	1.0	7–11
c'	1.3	0.2	1–2
V	88.3	1.3	86–91
M	82.0	2.6	76–86
G	59.7	6.8	48–70
Ovary, % of body length	43.7	5.9	36–53
Stylet, % of body length	21.8	2.3	17–26

<sup>a</sup> a = body length/greatest body width; b = body length/distance from anterior end to junction of esophagus and intestine; c = body length/tail length; c' = tail length/body width at vulva; V = distance of vulva from anterior end  $\times$  100/body length; M = conus length  $\times$  100/stylet length; G = distance from vulva to anterior end of ovary  $\times$  100/body length.

(1.0  $\pm$  0.1). Shaft diameter 1.0–1.6 (1.4  $\pm$  0.3). Stylet knobs rounded with concave anterior surfaces, 4.0–5.0 (4.6  $\pm$  0.4) wide, 6.0–20.0 (12.9  $\pm$  4.9) from guide ring. Procorpus strongly reflexed at level of metacarpus when stylet is retracted. Metacarpus ovoid, with a sclerotized valve slightly offset posteriorly. Nerve ring immediately posterior to metacarpus. Hemizonid position variable, 0–23.4 (10.4  $\pm$  8.4) posterior to nerve ring in gravid females. Intestinal contents usually yellow, depend-

ing on host diet, containing amorphous ingesta, crystalline inclusions, and bacteria. Intestine ending in a blind, rounded sac extending posterior to vulva. Esophageal glands 45.3–65.7 long (59.7  $\pm$  6.1) in small non-gravid females, 43.8–77.4 long (66.7  $\pm$  8.8) in large non-gravid females, and 62.8–81.8 (72.8  $\pm$  6.8) in large gravid females.

Ovary outstretched, rarely reflexed, frequently extending anterior to metacarpus in large specimens. Rachis present in large specimens. Oocytes progressively larger in posterior ovary, usually not more than one

TABLE 2. Measurements of 13 large paratype (>500  $\mu\text{m}$  body length), non-gravid female *Noctuidonema dibolia* n. sp. from Tift County, Georgia.

Character <sup>a</sup>	Mean	SD	Range
Measurements ( $\mu\text{m}$ )			
Body length	526	25.7	501–568
Maximum body width	57	6.6	44–67
Body width at nerve ring	36	4.6	28–44
Body width at vulva	44	6.7	32–54
Body width at anterior end of intestine	36	5.3	26–44
Anterior end to bulb base	125	13.6	96–143
Metacarpus length	28	2.8	20–31
Metacarpus width	24	1.9	20–26
Excretory pore to anterior end	19	2.5	15–23
Stylet total length	100	6.8	92–114
Stylet conus length	81	4.6	73–88
Head width	9	0.9	7–10
Head length	4	1.3	3–7
Ovary length	214	54.3	117–306
Vulva to end of tail	62	8.7	47–79
Vulva to anterior end	467	23.8	435–505
Vagina depth	27	5.4	15–34
Anterior end to anterior ovary	136	28.6	67–175
Ratios and Percentages			
a	9.3	1.0	8–11
b	4.2	0.4	4–5
c	8.7	1.1	7–11
c'	1.5	0.3	1–2
V	88.7	1.8	86–92
M	81.1	4.7	73–89
G	62.6	5.6	54–75
Ovary, % of body length	40.9	10.9	21–61
Stylet, % of body length	18.9	0.9	17–20

<sup>a</sup> a = body length/greatest body width; b = body length/distance from anterior end to junction of esophagus and intestine; c = body length/tail length; c' = tail length/body width at vulva; V = distance of vulva from anterior end  $\times$  100/body length; M = conus length  $\times$  100/stylet length; G = distance from vulva to anterior end of ovary  $\times$  100/body length.

TABLE 3. Measurements of 12 gravid paratype female *Noctuidonema dibolia* n. sp. from Tift County, Georgia.

Character <sup>a</sup>	Mean	SD	Range
Measurements ( $\mu\text{m}$ )			
Body length	541	54.4	451-640
Maximum body width	67	11.3	53-88
Body width at nerve ring	40	4.8	31-47
Body width at vulva	52	8.5	42-67
Body width at anterior end of intestine	40	4.8	31-47
Anterior end to bulb base	132	9.6	117-143
Metacarpus length	28	2.8	23-32
Metacarpus width	23	2.0	19-26
Excretory pore to anterior end	21	3.0	15-25
Stylet total length	102	7.0	91-114
Stylet cone length	82	3.3	76-86
Stylet shaft length	20	6.0	10-31
Head width	9	1.5	6-10
Head length	4	1.0	3-6
Ovary length	258	38.2	198-320
Vulva to end of tail	68	5.4	60-79
Vulva to anterior end	474	52.1	388-562
Vagina depth	30	7.3	20-45
Intrauterine egg length	86	8.5	66-98
Intrauterine egg width	30	2.5	26-35
Anterior end to anterior ovary	95	38.1	60-175
Ratios and Percentages			
a	8.2	1.1	7-10
b	4.1	0.3	4-5
c	8.0	0.7	7-10
c'	1.3	0.2	1-2
V	87.6	1.3	86-90
M	80.7	4.7	73-89
G	69.7	6.3	57-77
Ovary, % of body length	47.8	3.7	41-54
Stylet, % of body length	18.9	2.1	14-21

<sup>a</sup> a = body length/greatest body width; b = body length/distance from anterior end to junction of esophagus and intestine; c = body length/tail length; c' = tail length/body width at vulva; V = distance of vulva from anterior end  $\times$  100/body length; M = conus length  $\times$  100/stylet length; G = distance from vulva to anterior end of ovary  $\times$  100/body length.

mature egg in uterus. Bacteria frequently present, particularly in anterior ovary, and sometimes seen in intestinal epithelial cells, intestinal lumen, and eggs. Uterine glands not seen. Spermatheca ovoid, thick-walled, usually containing sperm. Intrauterine eggs  $30 \times 86 \mu\text{m}$ , usually in 1-8 cell stage.

Excretory pore 14-25  $\mu\text{m}$  from anterior end. Renette cell large, about  $20 \times 60 \mu\text{m}$ , consisting of a loosely coiled tubule extending anteriorly to the excretory pore

and posteriorly toward the tail. Renette cell nucleus large, 7.3-10.2 ( $8.8 \pm 1.2$ ) in gravid females, irregular in outline in fixed specimens, smooth in live specimens.

Tail shape conical, with mucro small or absent. Anus not visible with light microscopy. Vulva subterminal ( $V = 88\%$ ), transverse, with overlapping anterior lip. One pair of prevulval and one pair of postvulval papillae.

*Males:* Measurements in Table 4. Body hook-shaped, tapering gradually at ante-

TABLE 4. Measurements of 16 paratype male *Noctuidonema dibolia* n. sp. from Tift County, Georgia.

Character <sup>a</sup>	Mean	SD	Range
Measurements ( $\mu\text{m}$ )			
Body length	422	48.6	343-524
Maximum body width	42	3.1	38-51
Body width at nerve ring	31	2.5	28-35
Body width at cloaca	14	2.3	10-18
Body width at anterior end of intestine	31	2.5	28-35
Anterior end to base of bulb	106	14.0	77-126
Metacarpus length	26	1.5	23-28
Metacarpus width	20	2.1	16-23
Excretory pore to anterior end	21	4.2	15-31
Stylet total length	86	4.4	76-93
Stylet cone length	68	4.8	61-82
Stylet shaft length	18	5.1	16-26
Head width	8	1.0	7-10
Head length	4	0.9	3-6
Testis length	144	47.5	85-233
Spicule length	48	3.5	41-53
Spicule rostrum length	12	1.2	10-13
Cloaca to end of tail	9	1.1	6-10
Cloaca to anterior end	413	49.0	333-515
Anterior end to anterior testis	132	21.8	98-174
Ratios and Percentages			
a	10.0	1.1	8-12
b	4.0	0.3	3-5
c	50.8	11.1	34-80
c'	0.7	0.2	0-1
V	98.0	0.4	97-99
M	78.7	5.5	70-93
T	66.6	4.4	60-74
Testis, % of body length	34.6	12.1	21-56
Stylet, % of body length	20.7	2.1	17-24

<sup>a</sup> a = body length/greatest body width; b = body length/distance from anterior end to junction of esophagus and intestine; c = body length/tail length; c' = tail length/body width at cloaca; V = distance of cloaca from anterior end  $\times$  100/body length; M = conus length  $\times$  100/stylet length; T = distance from cloaca to anterior end of testis  $\times$  100/body length.

rior end, with posterior half strongly arcuate ventrally. Annules continuous, 1.2–1.8 wide ( $1.4 \pm 0.2$ ) at midbody, anastomoses common. Lateral field absent. Lip region low, bluntly rounded, not strongly set off, similar to female.

Stylet flexible, delicate, elongate, attenuated at tip, 17–24% of total body length. Conus representing about 79% of total stylet length. Stylet knobs rounded, with concave anterior surfaces. Stylet conus diameter 0.7–1.0 ( $0.9 \pm 0.1$ ), shaft diameter 1.0–1.6 ( $1.2 \pm 0.2$ ), knob width 2.6–5.0 ( $3.8 \pm 0.8$ ). Knobs to guide ring 6.8–17.0 ( $13.9 \pm 3.0$ ).

Metacarpus ovoid, with a sclerotized valvular apparatus offset slightly posteriorly. Procorpus strongly flexed at level of metacarpus. Esophageal glands 36.5–71.5 ( $58.1 \pm 11.38$ ) long, overlying anterior intestine. Nerve ring immediately posterior to metacarpus. Hemizonid position variable, 7.3–33.6 ( $15.9 \pm 8.5$ ) posterior to nerve ring. Intestine a blind sac, terminating well anterior to cloaca. Intestinal contents usually yellow, similar to female. Tail short, blunt, with one pair of precloacal and one pair of postcloacal papillae. Tail mucro absent or low. Spicules paired, sclerotized, nearly equal in size and shape, ventrally arcuate, with dorsal and ventral arms not well separated, ending in fine points closely apposed at tips. Matrix and sheath present, but greatly reduced. Rostrum slender, arcuate, distal end slightly enlarged. Manubria well developed, wedge-shaped, with right slightly more rounded than left. Gubernaculum and bursa absent. Cloacal cylinder thin-walled, projecting 4–6  $\mu\text{m}$  nearly perpendicular to tail axis during spicule protraction.

Testis single, outstretched anteriorly, without rachis. Anterior end of testis usually not extending anterior to metacarpus. Seminal vesicle containing spermatids and set off from remainder of testis by a moderate constriction. Bacteria commonly present in testis.

Excretory pore 15–30  $\mu\text{m}$  from anterior end. Renette cell large, about  $20 \times 50 \mu\text{m}$ ,

similar to female. Renette cell nucleus 4.4–10.2 ( $7.6 \pm 1.7$ ), similar to female.

#### *Type Host and Locality*

Ectoparasitic on adults of *Mocis latipes* (Guenée) (Lepidoptera: Noctuidae), particularly on intersegmental membranes of the abdomen. Type locality is the Bellflower Farm, located 5 km NNW of Tifton, Tift County, Georgia.

#### *Type Designations*

Holotype (female) (slide T-519t), allotype (male) (slide T-520t), 5 paratype slides (slides T-4526p–T-4530p), and one vial of paratypes in glycerin (vial T-370p) deposited in the U.S. Department of Agriculture Nematode Collection, Nematology Laboratory, USDA ARS, BARC-West, Beltsville, Maryland 20705. Four paratype slides deposited at each of the following: Museum National d'Histoire Naturelle, Laboratoire des Vers, Paris, France; Instituut voor Dierkunde, Gent, Belgium; Biosystematics Research Centre, Canadian National Collection of Nematodes, Canada Agriculture, Ottawa, Ontario, Canada; Rothamsted Experimental Station, Nematode Collection, Entomology and Nematology Department, Harpenden, Herts, United Kingdom.

#### *Diagnosis*

All stages of *N. dibolia* n. sp. are ectoparasitic on adult *Mocis latipes*. Stylet length is 83–114  $\mu\text{m}$  in females, 76–93  $\mu\text{m}$  in males, with a conus 73–89% of total stylet length in females and 70–93% in males. Cephalic end similar in males and females. Tail length is 42–79  $\mu\text{m}$  in females and 6–10  $\mu\text{m}$  in males. Spicules have dorsal and ventral arms closely apposed, not separated; the matrix and sheath are present but much reduced. A large renette cell is present in both sexes.

#### *Relationships*

Males of *N. dibolia* n. sp. are smaller and more slender than those of *N. guyanense*, have shorter stylets and shorter stylet conii,

and have spicules that differ in size and structure. The spicules of *N. dibolia* n. sp. are smaller and simpler than those of *N. guyanense* and have a greatly reduced matrix and sheath. In *N. dibolia* n. sp., the spicules have well-developed, wedge-shaped manubria, not reduced as in *V. daptria*, and have dorsal and ventral arms closely apposed, not widely separated as in *V. daptria*. Males of *N. dibolia* n. sp. are similar in size to *V. daptria* males, but have longer stylets and longer stylet conical.

Female *N. dibolia* n. sp. are smaller and more slender than those of *N. guyanense* and have a shorter stylet, shorter stylet cone, and longer tail. Female *N. dibolia* n. sp. differ from those of *V. daptria* in having a longer stylet, longer stylet cone, and shorter tail. The excretory pore is farther from the anterior end in *N. dibolia* than in *V. daptria*.

#### DISCUSSION

Hunt (7) erected the family Acugutturidae to include the genera *Acugutturus*, *Noctuidonema*, and the new genus *Vampyronema*. Remillet and Silvain (8) placed *Noctuidonema* in the subfamily Acugutturinae and distinguished it from *Acugutturus* by differences in body shape, stylet length, shape of head and tail, position of the excretory pore, shape and size of the spicules, and presence of a bursa. Anderson and Laumond (2) described *Noctuidonema daptria*, which Hunt (7) later redescribed as *Vampyronema* and placed with *Noctuidonema* in the newly created subfamily Noctuidonematinae.

Hunt (7) noted that basal swellings and knobs were absent on the stylet in *A. parasiticus*, and Remillet and Silvain (8) reported that stylet knobs were present in *N. guyanense*. Anderson and Laumond (1) provided more details on the morphology of the spicule and redescribed the male of *N. guyanense*. However, they also reported that the basal part of the stylet of *N. guyanense* is uniformly tubular, without swellings or knobs. Anderson and Laumond (2)

also reported the presence of stylet knobs in *N. daptria*. Stylet knobs are present in *N. dibolia*. We examined *Noctuidonema* specimens from several species of *Spodoptera* (*S. frugiperda*, *S. ornithogalli*, *S. latifascia*, and *S. androgea*) from the United States and French Guiana and found that stylet knobs were present in all *Noctuidonema* specimens examined. During processing of *N. dibolia* specimens for this report, we observed that the basal part of the stylet frequently becomes transparent, rendering the knobs difficult to see. However, the knobs are easily visible in living specimens. Our observations indicate that stylet knobs are present in all species of *Noctuidonema* and that this feature unites the known species of the subfamily Noctuidonematinae and aids in differentiating them from *Acugutturus*. Spicule structure in *N. guyanense* and *N. dibolia* are similar enough to justify grouping them within a single genus, and different enough to identify them as separate species and separate them from the related genus *Vampyronema*.

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