Pathological Effects of an Ectoparasitic Nematode Noctuidonema guyanense (Nematoda: Aphelenchoididae) on Adults of the Fall Armyworm (Lepidoptera: Noctuidae)

O. G. MARTI, C. E. ROGERS, J. F. SILVAIN,¹
AND A. M. SIMMONS

Insect Biology & Population Management Research Laboratory, USDA-ARS, Tifton, Georgia 31793

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ABSTRACT Male Spodoptera frugiperda (J. E. Smith) moths were collected in French Guiana and Martinique and examined for evidence of pathogenicity produced by natural infestations of Noctuidonema guyanense Remillet and Silvain, an ectoparasitic nematode. Nematodes may occur on the genitalia, any abdominal segment, or intersegmental membranes but are most often found on the anterior margin of the eighth tergite and the tergal lobes of the first abdominal segment. Stained sections and whole mounts of moth cuticle showed minor pathological changes attributable to nematode infestation. These changes included irregular dark spots 5–20 µm in diameter at the probable sites of stylet penetration, development of furrows occupied by nematodes, branched cuticular proliferations, minor cuticular erosion in the intersegmental spaces of the abdomen, and some evidence of scale loss. Hypertrophy of the intersegmental spaces of the abdomen occurs in heavy infestations. The acute effects of feeding by N. guyanense on the energy reserves, flight ability, or fecundity of its hosts were not determined.

KEY WORDS Insecta, nematoda, parasite, pathology, Spodoptera frugiperda

Noctuidonema guyanense Remillet and Silvain was described from noctuid moths of the genera Spodoptera, Anicla, and Leucania in French Guiana (Remillet & Silvain 1988). Rogers et al. (1990) found that N. guyanense occurred in 25 species of Noctuidae, including several species of Spodoptera and Mocis, in French Guiana. Recently, N. guyanense has also been found to be a common parasite of the fall armyworm, S. frugiperda (J. E. Smith), and several other noctuid species trapped in Georgia, Florida, and countries of the Caribbean Basin (A.M.S. et al., unpublished data).

Noctuidonema guyanense is unusual in that it is ectoparasitic, occurring primarily on the intersegmental membranes of the abdomen of both sexes of its adult hosts. Although details of its life cycle are unknown, Remillet & Silvain (1988) reported that all stages of the parasite occur on the moth, that infestation may be transmitted by mating, and that the parasite is not found on the larvae of host species. A related ectoparasitic nematode, Acugutturus parasiticus Hunt, was described from the cockroach, Periplaneta americana (L.), in St. Lucia, West Indies (Hunt 1980).

Parasitoids and other natural enemies are very important in the control of *S. frugiperda* and other pest insects of agriculture. Ashley (1979) listed 53

species of parasitic Hymenoptera and Diptera reared from larvae of S. frugiperda. However, no parasitoids are known to attack the fall armyworm adult. With the exception of incidental or experimental infestation by steinernematid and heterorhabditid nematodes (Triggiani & Poinar 1976, Kaya & Grieve 1982, Timper et al. 1988), mites (Treat 1969) and mermithid nematodes (Welch 1963) are the only metazoan parasites previously reported from adult Lepidoptera. There are no current biocontrol programs against adult S. frugiperda because suitable parasites or pathogens of the adults are not known. As a parasite of the adult, N. guyanense may shorten the life span or reduce the flight ability and fecundity of its host. In addition, the presence of moths bearing N. guyanense in northern regions of the United States may provide information of value in the study of the migratory habits of S. frugiperda.

This study was conducted to determine the pathological effects of natural infestations of *N. guyanense* on adult male *S. frugiperda*. For the purposes of this report, we assume that *Noctuidonema* in North American *S. frugiperda* is conspecific with *N. guyanense* as described by Remillet & Silvain (1988). However, additional study is required to confirm this.

Materials and Methods

Adult male S. frugiperda were captured in universal moth traps baited with a commercial formulation of S. frugiperda sex pheromone. Traps

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¹ Formerly of Centre ORSTOM, Cayenne, Guyane Française; currently at INRA, Station de Recherches de Lutte Biologique, La Minière, 78280, Guyancourt, France.

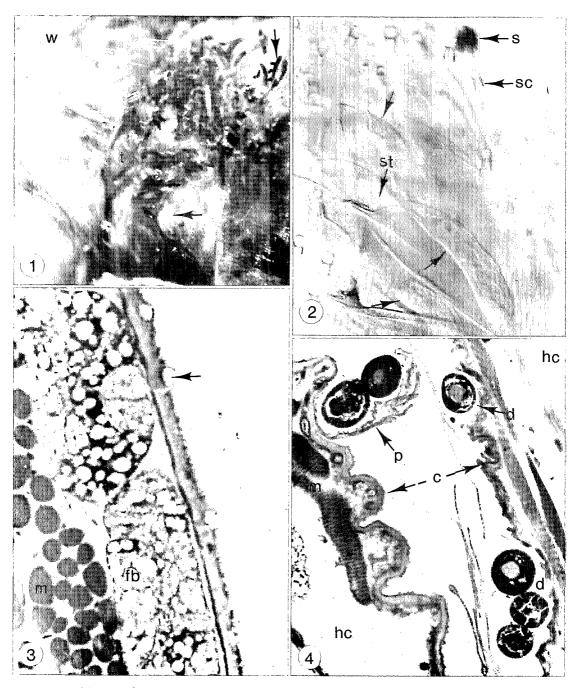


Fig. 1–4. (1) Noctuidonema-infested S. frugiperda from Tift County, Ga., 1988; base of hindwing (w), tympanic opening (t), Noctuidonema guyanense in situ (arrows), $24\times$. (2) Cuticle from eighth tergite of N. guyanense-infected S. frugiperda from Martinique, W.I., with dark feeding site (s), scale socket (sc), stylet (st), and larval N. guyanense (arrows), $407\times$. (3) Cross-section of abdomen from a laboratory-reared S. frugiperda; muscle (m), fat body (fb), cuticular pit (arrow), $407\times$. (4) Cross-section through abdominal intersegmental space of Noctuidonema-infested S. frugiperda from French Guiana; hemocoel (he), muscle (m), cuticle (c), cuticular projection (p), cuticular depressions with N. guyanense cross-sections (d), $403\times$.

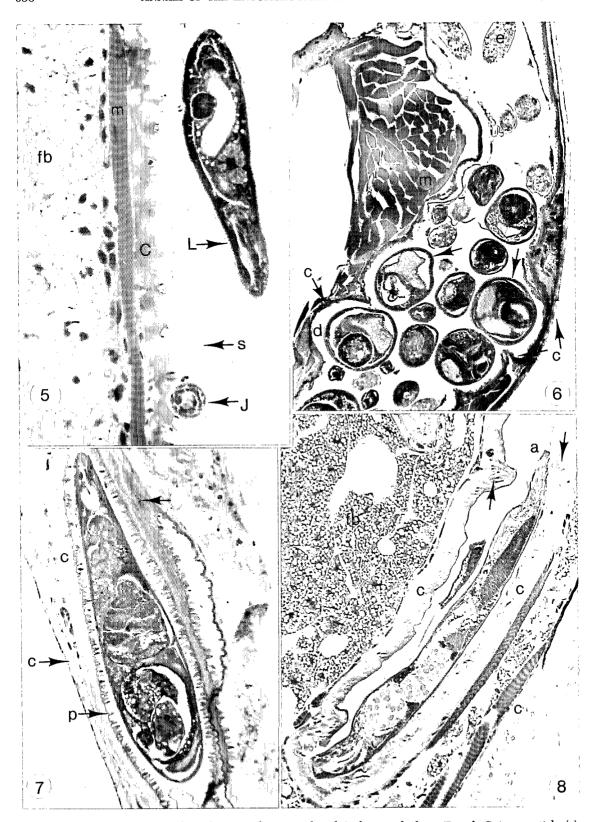


Fig. 5–8. (5) Cross-section through *Noctuidonema*-infested *S. frugiperda* from French Guiana; cuticle (c), scales (s), muscle (m), fat body (fb), cross-section of juvenile (J), longitudinal section of juvenile (L), 415×. (6) Cross-section through intersegmental space of *Noctuidonema*-infested *S. frugiperda* from Martinique, W.I.; cuticle (c),

were placed near Matoury, French Guiana (the *N. guyanense* type locality) in 1987, on Martinique in 1988, and near Tifton, Ga., in 1988.

Abdomens from freshly collected moths were removed, fixed in 10% formalin, and stored until examination. Male S. frugiperda obtained from the Insect Biology and Population Management Research Laboratory (IBPMRL) colony were used as uninfested controls. Abdomens selected for sectioning were dehydrated through ethanol (30-100%) and infiltrated, oriented, and embedded in Sorvall embedding medium (E. I. du Pont de Nemours & Company, Wilmington, Del.). Transverse and longitudinal sections were cut at 2-6 µm with hand-broken glass Ralph knives. Sections were stained in borate-buffered 0.5% Toluidine blue (E. F. Fullam, Latham, N.Y.) for 1-3 min, rinsed in water, cleared in xylene, and mounted in Permount (Fisher Scientific Company, Fairlawn, N.J.). In some specimens, portions of the posterior cuticle were dissected, dehydrated, cleared, and mounted without staining directly into Permount as whole mounts. Slides of sections or whole mounts were examined with a Nikon Optiphot photomicroscope using brightfield illumination or Nomarski differential interference contrast optics and photographed with Kodak Technical Pan film (Eastman Kodak, Rochester, N.Y.).

Results and Discussion

Remillet & Silvain (1988) reported that N. guyanense were found on posterior segments of S. frugiperda abdomens. In our study, the nematodes were found more frequently on the eighth tergite than elsewhere. Here, they were usually oriented with their heads near, or in contact with, the membrane between the eighth and seventh segments. Other sites often bearing nematodes were the claspers, the genital chamber, the tergal lobes of the first abdominal segment (Fig. 1), and occasionally the thorax. Newly hatched juveniles, often in groups, were present beneath scales of infested moths (Fig. 2). With the possible exception of scale loss, no lesions specifically attributable to juveniles could be detected by light microscopy.

Examination of S. frugiperda abdomens revealed a variety of pathological effects attributable to infestation by N. guyanense. Approximately 85% of infested moths captured in Tift County, Ga., in 1988, had small, irregular dark spots (10–50 μ m in diameter) on the cuticle (Fig. 2). These spots were generally limited to the posterior abdominal cuticle and to the intersegmental membranes associated with abdominal segments seven and eight. Spots



Fig. 9. Longitudinal and cross-sections of adult males (m), adult females (f), and eggs (e) of *N. guyanense* under host-derived membrane (arrows) on *S. frugiperda* from Martinique, W.I., 99×.

were not seen on uninfested wild moths nor on moths reared from the IBPMRL colony. Small spots varied in color from yellow to light brown, whereas larger spots were usually brown to black. The spots may represent nematode feeding sites that become darkened with oxidized hemolymph after the nematodes retract their stylets and move elsewhere. The actual stylet penetration sites were not observed because they were obscured by accumulated oxidized material composing a spot. The cuticle of infested moths was generally smooth or revealed only scale sockets or small pits with sharply defined margins (Fig. 3).

The presence of spots on the tergites suggests that *N. guyanense* can feed through the cuticle as well as through the thinner intersegmental membranes. Hunt (1980) reported that feeding of a related nematode, *A. parasiticus*, was limited to host hemolymph. Electron microscopy may be required to explain details of stylet penetration and

intersegmental space with cuticular depressions (d) containing cross-sections of *N. guyanense* (arrows), 205×. (7) Longitudinal section of adult female *N. guyanense* in intersegmental space of *S. frugiperda* from French Guiana; cuticle (c), cuticular projections (p), possible stylet penetration site (arrow), 200×. (8) Longitudinal section of adult male *N. guyanense* in intersegmental space of *S. frugiperda* from French Guiana; cuticle (c), fat body (fb), anterior end of nematode (a), possible feeding sites (arrows), 225×.

to determine whether *N. guyanense* feeds on hemolymph, cell solutes, or upon specific host tissues.

Cross-sections of moth abdomens showed that the nematodes often lie in cuticular depressions or furrows approximately the same size and shape as the nematode (Fig. 4–6). The cuticle and hypodermis were not thinned or eroded by the nematodes but were shaped to accommodate their presence. The depressions varied from broad and shallow furrows (Fig. 4) to well-defined hemispherical cavities (Fig. 6). These cuticular depressions are sharply defined when many nematodes occur together, particularly in the apices of the intersegmental spaces (Fig. 6).

Long, branched extensions of host cuticle in the intersegmental spaces, while not common, were usually associated with juvenile nematodes (Fig. 4). Short, unbranched cuticular processes in the intersegmental space were sometimes associated with the presence of one or more adult nematodes (Fig. 7). Because these latter processes were seen in infested and uninfested moths, they must be normal cuticular structures of the intersegmental space, although the opposite impression might be obtained by studying only infested moths. Minor localized erosion of cuticle in the intersegmental spaces also was associated with adult nematodes (Fig. 8). When many nematodes were present, the intersegmental space was greatly enlarged at the expense of the hemocoel (Fig. 6). In some heavily infested moths, nematodes and eggs may be found beneath membranous tissue or exudate of host origin, where they are difficult to see and are easily overlooked (Fig. 9). Bacteria were seen in association with living and dead nematodes; however, there was no indication that the bacteria were pathogenic to either the nematodes or the moth.

Results of this study confirm that *N. guyanense* is parasitic, as reported by Remillet & Silvain (1988), and not merely commensal or phoretic. All *S. fru-giperda* specimens examined in this study were capable of flight because they were captured in pheromone traps. Additional pathological changes may occur in severely infested individuals with impaired flight. Although only male *S. frugiperda*

were examined in this study, we have no reason to believe that the pathological changes reported here are unique to male hosts.

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