OBSERVATIONS ON A PARASITIC NEMATODE (TETRADONEMATIDAE) OF FIRE ANTS, SOLENOPSIS (FORMICIDAE), FROM MATO GROSSO¹

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ABSTRACT - Parasitic nematodes, *Tetradonema* sp. (Mermithoidea: Tetradonematidae) were observed in the fire ant, *Solenopsis invicta* Buren, for the first time. Five colonies in a sample of 14 colonies from one site in Mato Grosso were infected. The infection rate among adult workers was 12.5% in one colony and less than 5% in each of the remaining colonies. Adult reproductive males and females, eggs, and juvenile nematodes were observed in the haemocoel of male and worker ants. The infected worker ants often can be recognized by their slightly enlarged gasters, the dorsal sclerites of which may have a scalloped appearance. There are no other morphological signs of infection and no observable changes in behavior. Infected adult males contained normal sperm. An attempt to transmit the nematode to colonies of *S. invicta* in the United States was unsuccessful.

Index terms: Solenopsis invicta, Tetradonema, gaster, sclerites, morphology, infection.

OBSERVAÇÕES SOBRE UM NEMATÓIDE PARASITO (TETRADONEMATIDAE) DA FORMIGA LAVA-PÉ, SOLENOPSIS (FORMICIDAE), EM MATO GROSSO

RESUMO - Foi observado pela primeira vez um nematóide da família Tetradonematidae parasitando Solenopsis invicta Buren. Cinco colônias de um total de quatorze coletadas em Mato Grosso estavam infectadas. A taxa de infecção de adultos foi de 12,5% em uma colônia e as outras colônias restantes apresentaram menos de 5%. As formigas infectadas podem ser reconhecidas por apresentarem os gásteres ligeiramente maiores e os escleritos dorsais elevados. Não existem outros sinais morfológicos de infecção e nem modificações no comportamento que indiquem a infecção. Os machos adultos infectados tinham esperma normal. A tentativa de transmissão do nematóide para colônias de *S. invicta* nos Estados Unidos da América foi negativa.

Termos para indexação: Solenopsis invicta, Tetradonema, infecção, gáster, escleritos, morfologia.

INTRODUCTION

Two species of fire ants, Solenopsis richteri Forel and Solenopsis invicta Buren, were introduced into the United States from South America. 1918 and 1940, respectively (Buren 1972). These ants now infest over 10^8 ha in nine south-eastern states, and are slowly spreading. If they are transported by man across the barrier of the arid southwestern United States to more humid or irrigated areas, their range could greatly increase.

S. invicta, known in the United States as the "red imported fire ant", occupies at least 95% of

the currently infested area. Except for one small enclave, S. richteri, the "black imported fire ant," has been displaced by the more recently introduced species. The native range of S. invicta is west-central Prazil (Rondonia, Mato Grosso, Mato Grosso do Sul) south along the Paraguay River into northern Argentina. S. richteri is native to Uruguay and parts of Argentina and southern Brazil (Buren et al. 1974).

The imported fire ants are medically and agriculturally important pests in the United States (Lofgren & Adams 1982). Because of their ability to dominate the ant fauna in disturbed areas and because of concerns about the use of pesticides to control them, there has been intense interest in the United States in the possibility of controlling them biologically. In the United States they are essentially free of natural enemies (Jouvenaz et al. 1977); in Brazil, they are beset by both diseases and arthropod enemies (Jouvenaz 1983). Therefore, the Insects Affecting Man and Animals Re-

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search Laboratory, Agricultural Research Service (ARS), U.S. Department of Agriculture (USDA), concluded a cooperative agreement with the Empresa Brasileira de Pesquisa Agropecuária (EMBRAPA) in September 1983, to survey for natural enemies of fire ants, and also to explore means of identifying the natural enemies of leafcutting ants. The goal of this research is to discover and evaluate specific parasites and diseases of fire ants. Natural enemies potentially useful for fire ant control will be considered for introduction into the United States.

The nematode reported here is the first to be found in fire ants in South America. The only other nematode reported from fire ants is an unidentified (but different) species that was found recently in a few alcohol-preserved specimens of *Solenopsis geminata* (F.), a species native to Florida (Mitchell & Jouvenaz 1985).

MATERIAL AND METHODS

The procedures for screening large numbers of ants for pathogens have been described by Jouvenaz et al. (1977). Samples of soil are taken from nests into the laboratory in small buckets, the sides of which have been dusted with talc to prevent the ants from escaping. The soil is moistened if necessary, and allowed to stand undisturbed overnight to allow the ants to establish tunnels and gather the brood. The ants are forced to the surface by slowly dripping water into the soil until it is completely submerged. The ants (including immatures rescued by the adult workers) float in masses and are transferred easily to new containers. A sample of ca. 1,000 to 2,000 mixed adults and immatures from each colony is then triturated in distilled water in a glass tissue grinder, and the crude extract is examined by phase-contrast microscopy for fungal mycelia or spores, protozoan spores, virus polyhedra, nematode eggs or larvae, and unusual numbers of bacteria. Non-occluded viruses cannot, of course, be detected by this procedure. When extracts containing suspected pathogens are found, individual ants from the colony are examined to determine whether disease is indeed present.

Nematode eggs, juveniles, and adults dissected from the gasters of adult worker ants were preserved in 3% formalin containing 2% glycerine (v/v). Measurements were made from calibrated phase-contrast photomicrographs. The intracolonial infection rates were determined by phase-microscopic examination of individual squashes of 200 workers randomly selected. In the most heavily infected colony, 200 worker pupae and 200 adult males, and all 65 adult alate females in the collection, were also examined, as were three adult and fourteen pupae of the

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parasitic wasp, *Orasema* sp. (Hymenoptera: Eucharitidae). Adult worker ants from each colony were preserved in 70% ethanol for positive identification and for examination for morphological aberrations.

An attempt was made to infect two small colonies of *S. invicta* from Florida with the nematode. Workers from the most heavily infected colony were hand-carried (under Brazilian and USDA permits) to the Insects Affecting Man and Animals Research Laboratory, ARS, USDA, Gainesville, Florida, USA. There they were maintained on autoclaved soil in two large petri dishes for three weeks, then removed from the soil and replaced with two small colonies of *S. invicta*. After two months, the *S. invicta* colonies from Florida were examined for infection. Living and preserved workers were also submitted to Dr. W.R. Nickle, Beltsville Agricultural Research Center, ARS, USDA, Beltsville, Maryland.

RESULTS

A nematode, Tetradonema sp. (Mermithoidea: Tetradonematidae), infected five of fourteen colonies of S. invicta collected 5 February 1985, from a 250 - m section of roadside at km 616, BR 070, between Cuiabá and Cáceres, MT (disturbed cerrado). In the most heavily infected colony, 12.5%. of the adult workers and 2% of the adult males were infected. None of the 65 adult females collected from this colony were infected, nor was infection detected in worker pupae (early infection may be difficult to detect). The specimens of parasitic wasps, Orasema sp., from this colony were not infected. In the other four colonies, the infection rates were no more than 5% in adult workers.

The collection site for the infected colonies was a roadside having a slope ca. $15^{\circ} - 20^{\circ}$. No pattern of distribution of infected colonies was apparent, as non-infected colonies were located between infected colonies. Two infected colonies were located upslope (34 m and 53 m) and two downslope (108 m and 119 m) from the colony with the highest infection site. All of the colonies, including the infected ones, were large and contained abundant brood.

Eggs, juveniles, and egg-laying adult nematodes were found in *S. invicta* workers. The eggs and juveniles (Fig. 1) are readily observed in aqueous extracts or squashes of ants (phase-contrast microscopy). By placing a living ant in water under



FIG. 1. Two nematode eggs and a juvenile from the fire ant, *S. invicta.* X 650.

a cover slip, the active nematode juveniles may also be seen under low magnification (200 X) phase-contrast microscopy in the haemolymph throughout the body of the host, including the mandibles, antennae, and tarsi. The eggs and adults are confined to the gaster. The adult nematodes are usually destroyed in extracts and squashes, and are best observed by dissecting the gasters of large workers (Fig. 2 and 3).

The adult nematodes measure 1.2 mm to 1.4 mm in length and ca. 0.17 mm in diameter. From 17 to 35 were dissected from the gasters of large workers. These rather thick-bodied nematodes contained eggs throughout their bodies. The juveniles measure ca. 0.15 mm in length, the eggs slightly less than 0.04 mm in diameter. At least 1,000 eggs and juveniles (total) were present in large workers. W.R. Nickle, U.S.D.A., ARS, Beltsville, MD,



FIG. 2. Adult female nematodes in a partially dissected gaster of a worker fire ant, *S. invicta*. X 32.



FIG. 3. Adult female nematodes dissected from the gaster of a worker fire ant, *S. invicta*. Divisions are in mm. X 15.

observed a small adult male nematode in a worker ant.

Living ants parasitized by these nematodes may often be recognized by their slightly enlarged gasters, the dorsal sclerites of which have a scalloped appearance. There are no other morphological signs of infection, nor are there any observable changes in behavior. All four of the infected males contained sperm. An attempt to transmit the infection to two colonies of *S. invicta* from Florida was not successful.

The nematode was collected in February 1985. In May, W.A. Banks, D.F. Williams, and A.C.C. Pereira screened eight colonies from the roadsides adjacent to both ends of the collection site but did not find the infection. In June 1984, D.P. Wojcik had collected this same site without finding nematodes.

DISCUSSION

Were examined over 1,200 colonies of fire ants from a 150 km radius of Cuiabá, and south through Rondonopólis, Coxim, and Campo Grande. Several pathogens have been observed commonly (Jouvenaz 1983). The presence of nematodes in five colonies (in a collection of 14 colonies) from one site on one occasion indicates that this parasite may be unevenly distributed, or cyclic, or both. The failure to transmit the infection in the laboratory may possible be due to factors that restrict the natural distribution of the nematode. The propagation and transmission, life cycle,

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host range and pathology, and classification of this potential biological control agent will be the subject of future research.

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REFERENCES

BUREN, W.F. Revisionary studies on the taxonomy of the imported fire ants. J. Ga. Entomol. Soc., 7:1-26, 1972.

- BUREN, W.F.; ALLEN, G.E.; WHITCOMB, W.H.; LEN-NARTZ, F.E.; WILLIAMS, R.N. Zoogeography of the imported fire ants. J.N.Y. Entomol. Soc., 82: 113-24, 1974.
- JOUVENAZ, D.P. Natural enemies of fire ants. Fla. Entomol., 66:111-21, 1983.
- JOUVENAZ, D.P.; ALLEN, G.E.; BANKS, W.A.; WOJ-CIK, D.P. A survey for pathogens of fire ants, Solenopsis spp., in the southeastern United States. Fla. Entomol., 60:275-9, 1977.
- LOFGREN, C.S. & ADAMS, C.T. Economic aspects of the imported fire ant in the United States. In: BREED, M.D.; MICHENER, C.D.; EVANS, H.E., ed. The biology of social insects. Boulder, Westview, 1982. 419p.
- MITCHELL, G.B. & JOUVENAZ, D.P. Parasitic nematode observed in the tropical fire ant, Solenopsis geminata (F.) (Hymenoptera: Formicidae). Fla. Entomol., 68:492-3, 1985.