Reprinted from *Florida Entomologist*, Volume 73, Number (1), March, 1990.

STEINERNEMATID NEMATODE DRENCHES FOR CONTROL OF FIRE ANTS, *SOLENOPSIS INVICTA*, IN FLORIDA

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Recent developments in low-cost, large-scale production of entomogenous nematodes of the genera *Steinernema* and *Heterorhabditis* (Bedding 1984) have facilitated large-scale field testing against various soil-infesting insect pests (Kaya 1985, Poinar 1986), with good potential for commercial control in some applications. Early research by Poole (1976) and Quattlebaum (1980) showed promise that entomogenous nematodes have potential for controlling fire ants, *Solenopsis* spp. Furthemore, these nematodes have the ability to invade adult insects, and thus potential to kill fire ant queens. Agents which only kill worker ants do not effectively eliminate colonies, although they may temporarily reduce their size.

A variety of entomopathogens, including viruses, bacteria, fungi, protozoa, and other species of nematodes have been evaluated for fire ant control. These organisms, with the possible exception of a fungus (*Beauveria* sp.) isolated from fire ants in Brazil (Stimac et al. 1987), produced little or no mortality in laboratory and/or field tests. Jouvenaz (in press) recently reviewed these evaluations and discussed the special problems attending the development of biological formicides.

We selected a strain of *Steinernema feltiae* produced by Biosys Inc. (Palo Alto, CA) for field evaluation after screening several strains of *Steinernema* and *Heterorhabditis* spp. in the laboratory. In the laboratory tests, groups of ants composed of queens (from a polygynous colony), about 25-30 workers, and a small quantity of brood were placed in 16 x 125 mm glass culture tubes containing 25 mm of slightly moistened sandy soil. The tubes were capped with plastic tops which allowed gas exchange and were maintained at 29° C. After two or three days of acclimation, one ml of deionized water without nematodes or containing 5,000 or 50,000 nematodes was added to each tube, and survival of queens was scored daily for four days. Ten replicates (a total of 50 queens) were used for each nematode concentration and for the control. After 96 h, the strain we selected had produced queen mortalities of 40% and 58% in 5,000 and 50,000 nematode dosaged, respectively. Mortality of workers and brood, although not quantitatively determined, was estimated to be at least 80%. Mortality of queens, workers or brood was not observed in the control group. In separate tests we observed that colonies of ants vacated soil treated with high concentrations of nematodes.

The first of the two field evaluations reported here was conducted in October and November, 1987, at Site No. 1 in southeast Gainesville, Alachua Co., Florida. This site was a utiities right-of-way measuring about 365 x 23-30 m, with sandy soil and surrounded by trees. The site had been mowed regularly prior to the test, but was not mowed during the test period. The second test was conducted during June and July, 1988, at Site No. 2, 4 mi north of LaCrosse, Alachua Co., Florida. This site was an approximately rectangular plot 1.4 ha in size on a sod farm and was also bordered by trees. On both plots, the grass varied from 10-30 cm high, and was not mowed during the test period.

Site no. 1 was divided crosswise into four areas of approximately equal size (about 90 x 23-30 m); mounds on the site (including abandoned mounds) were marked with wooden stakes and color-coded according to treatment with flagging ribbon. The active mounds were rated by the population index method of Williams & Lofgren (1983) before treatment and at intervals of 7, 14, 28, and 45 days after treatment. Abandoned mounds were also checked to determine whether treated colonies had moved into them. Four treatments (one treatment per plot) were used in which all mounds in the plot were either: 1) drenched with one gallon of water; 2) drenched with one gallon of water containing 5 x 10^6 infective juvenile nematodes (the viability of the nematodes was verified microscopically within 1 h before they were transported to the field in styrofoam chests containing ice); 3) treated with 5 tablespoons of Amdro fire ant bait according to the label recommendations, or 4) received no treatment. The mounds were examined with minimal disturbance several times weekly to monitor colony movement. No rainfall occurred during the first 28 days of the test. The results of this test are presented in Table 1.

Site no. 2 was divided into 12 plots which were randomly assigned one of three treatments: 1) 2 x 10^6 infective juvenile nematodes (dessicated product) in one gallon of water; 2) Amdro fire ant bait; or 3) no treament. All mounds were marked with wire survey flags, color coded for treatment, and rated as at Site No. 1 prior to treatment and 40 days after treatment. During the interim, each mound was checked 48 hours after treatment and at approximately weekly intervals thereafter by stamping the ground nearby to disturb the ants and thereby verify that the mound was still active. If ants did not appear from this disturbance, a search for new, unmarked mounds in the vicinity (up to 10 m) was made to determine whether the colony had moved. The results of this test are presented in Table 2.

Although the strain of *S. feltiae* used in these tests killed up to 58% of queens exposed in the laboratory and also produced very high mortality in adult and immature workers, it was not effective against colonies in either field test. In the first test (Site No. 1), we suspected that very dry soil affected the nematodes adversely. The increase in population index of the non-treated colonies 45 days after treatment (Table 1) followed a rain which occurred 30 days after treatment. The Amdro treatment was significantly different (Waller-Duncan K-Ratio t-Test; PC-SAS 6.03, SAS Institute Inc., Cary, NC) from all other treatments, whereas there was no significant difference ($p = 0.05$) between the water and nematode treatments, or between the untreated and nematode treated. At least one inch of rain fell during each week of the second test; however, there was no difference in population index reduction between the nematode treated

TABLE 1. REDUCTION IN POPULATION INDICES (P.I.) OF IMPORTED FIRE ANT MOUNDS AT SITE #1.

Treatment	No. Active Mounds	Initial P.I.	P.I. Reduction (%) Days after treatment			
			7	14	28	45
Amdro[1] A[2]	25	367	25.1	46.6	60.5	65.1
Water[3] B	21	339	10.0	18.0	20.9	23.6
Nematode[4] BC	19	292	10.6	25.7	31.5	7.9
Untreated C	18	353	10.5	17.3	15.0	–15.1

[1]5 Tablespoons per mound.
[2]Treatments followed by the same letter are not significantly different. Regardless of treatment, there are no significant differences over time (Waller-Duncan K-Rtio t-Test).
[3]1 gallon of water.
[4]5x10^6 *S. feltiae* per mound in 1 gallon of water.

TABLE 2. REDUCTION OF POPULATION INDICES (P.I.) AND ACTIVE MOUNDS OF IMPORTED FIRE ANTS AT SITE NO. 2, 40 DAYS AFTER TREATMENT. DATA ARE SUMS OF FOUR PLOTS FOR EACH TREATMENT.

	Before Treatment		After Treatment	
Treatment	# Active Mounds	Initial P.I.	# P.I. Reduction After Treatment	% Reduction Active Mounds
Amdro[1]	52	624	96.5%	92.3
Nematode[2]	44	588	15.0%	27.3
Untreated	49	743	6.1%	12.2

[1]5 Tablespoons per mound.
[2]5 x 10^6 *S. feltiae* per mound in 1 gallon of water.

and untreated plots (the population index increased on one of the untreated plots, but the reduction in the remaining three plots averaged 10.9% vs. 15.0% for the nematode plots).

Quattlebaum (1980) reported mound mortalities of 88.2 to 96.8% using 2 x 10^6 *Neoaplectana carpocapsae* ("DD-136") per mound, and 75 to 86.3% using *Heterorhabditis heliothidis* ("NC-19") in South Carolina. Poole (1976) reported mound mortalities of 35% (fall treatment) and 45% (spring treatment) using 10^6 *Neoaplectana dutkyi* per mound in Mississippi. Their criteria for evaluating treatments differed from ours, however, and they apparently did not consider mound movement. Poole (1976) reports spading up mounds and spraying the workers with paint to identify them and of destroying "new migratory colonies". If mound movement is ignored (all treated and control mounds uninhabited after treatment are scored as dead), control by *S. feltiae* appears to be ca. 78% in our second test. These investigators also applied treatments under more favorable conditions such as during early morning or late evening precipitation.

Colony movement was stimulated by the presence of nematodes. After 72 h, 10 (52.6%) of the 19 colonies treated with nematodes in the first test had moved, whereas only four (16.7%) of the 24 colonies treated with Amdro, five (23.8%) of the 21 colonies treated with water, and none of the untreated colonies moved. In the second test, stimulation of colony movement by nematodes was even more pronounced. After 48 h, 27 (61.3%) of the 44 colonies treated with nematodes had moved, whereas only four (7.6%) of the 52 colonies treated with Amdro, and five of the 49 colonies (10.2%) of the control colonies had moved.

S. feltiae applied to dry sandy soil may lose activity before infecting insect hosts (Molyneux & Bedding 1984). Even with adequate soil moisture, the pathogenic lifespan of entomogenous nematode is limited by the ambient microfauna present in field soils (Ishibashi & Kondo 1986, 1987). Thus, any attempt to control fire ants with entomogenous nematodes will need to overcome these disadvantages either through selection of a superior strain or through improved formulation of existing strains. Efficacy of drench treatments of fire ants may always be limited by the tendency of colonies to relocate some distance from the antagonistic agent. Thus, alternative application strategies such as uniform application (through a sprinkler or irrigation system, for example) to create areas void of fire ants, bait traps containing nematodes, or slow release formulations in hygroscopic polymers (Schroeder, in press) may offer improvements in efficacy.

We thank Mr. Lloyd R. Davis, Jr. for excellent technical assistance, and an anonymous reviewer of the manuscript for many helpful suggestions. Mention of a commercial or proprietary product does not constitute an endorsement of that product by the U.S. Department of Agriculture.

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