RESEARCH NOTE/NOTA DE INVESTIGACIÓN

REPRODUCTIVE RATE DIFFERENCES OF ROOT-KNOT NEMATODES FROM MULTIPLE CROPS IN A SINGLE FIELD

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ABSTRACT

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Three soil samples were taken from a root-knot nematode (*Meloidogyne,* RKN) infested field in central Alabama at the Auburn University Plant Breeding Unit (PBU) near Tallassee, AL, in October of 2016. Each soil sample represented the crop planted in the field: cotton, soybean, and corn. A differential-host test was run on each of the samples for RKN species and race identification, and for host range and reproductive analysis. The reproductive factor was calculated for each population on eight different crops, and all three samples were determined to be *Meloidogyne incognita* race 3. Reproductive factors for each population varied across the different hosts, but all three populations were the same RKN species and race. Thus, management for each section of the field remains the same in regard to crop rotation and nematicide application. However, these results do suggest that there is some variability of the reproductive factor among host crops despite being the same species and race of RKN.

Key words: Cotton, corn, crop rotation, nematode management, *Meloidogyne* spp., root-knot nematode, soybean

RESUMEN

Groover, W., K. S. Lawrence, and P. Donald. 2019. Diferencias en la tasa de reproducción de los nematodos del nudo de la la raíz de múltiples cultivos en un solo campo. Nematropica 49:152-156.

Se tomaron tres grandes muestras de suelo de un campo infestado de nematodos de nudo de raíz (*Meloidogyne*, RKN) en el centro de Alabama en la Unidad de Fitomejoramiento (PBU) de la Universidad de Auburn cerca de Tallassee, AL, en octubre de 2016. Cada muestra de suelo representaba el cultivo plantado en el campo: algodón, soja y maíz. Luego se realizó una prueba de huésped diferencial en cada una de las muestras para la identificación de especies y razas RKN, y para el análisis de rango y reproducción del huésped. El factor reproductivo se calculó para cada población en ocho cultivos diferentes, y se determinó que las tres muestras eran *Meloidogyne incognita* raza 3. Hubo cierta variabilidad en los factores reproductivos para cada población en los diferentes huéspedes, pero las tres poblaciones eran las mismas RKN especie y raza. Por lo tanto, el manejo para cada sección del campo sigue siendo el mismo con respecto a la rotación de cultivos y la aplicación de nematicidas. Sin embargo, esto sugiere que puede haber cierta variabilidad del factor reproductivo entre los cultivos hospedantes originales a pesar de ser la misma especie y raza de RKN.

Palabras clave: Algodón, maíz, manejo de nematodos, *Meloidogyne* spp., Nematodo de nudo de raíz, rotación de cultivos, soya

The root-knot nematode (RKN, *Meloidogyne* spp.) is one of the most damaging plant-parasitic nematodes in the world. It has been estimated to account for 14.6% of crop loss in tropical and sub-tropical climates, and 8.8% in developed countries (Nicol *et al*., 2011). With such a high potential for crop damage, detection and management of RKN populations in a field are very important. Since all species of RKN are host dependent, a grower can sometimes rotate to a nonhost crop if the nematode is identified to species level. For example, cotton growers often have infestations of *M. incognita*. To help lower RKN population density, a common crop rotation is from cotton to peanut. However, peanut is an excellent host of *M. arenaria*, which can be found in the same field as *M. incognita* (Davis and Timper, 2000). Thus, knowing what species is present in a field is key for RKN management.

One of the most common methods to identify RKN species is through the differential-host test. This test was originally developed by Sasser (1972) to determine the species and race of four commonly found plant-parasitic root-knot nematodes in the southeastern United States. These are *M. incognita* (race 1-4), *M. arenaria* (race 1-2), *M. javanica,* and *M. hapla.* For the differential-host test (DHT), an unknown nematode population is inoculated onto cotton (Deltapine 16), tobacco (NC 95), pepper (California Wonder), watermelon (Charleston Grey), peanut (Flor-runner), and tomato (Rutgers). After 45 days, the plants are harvested and host determination is made of each crop for the population, allowing for species and race identification to be made based upon the RKN populations reproductive host range. The overall objective of this study was to determine if reproductive factors calculated from the DHT of a RKN population differ based on the host crop in the field.

Soil samples were taken from a 1.5-ha area section of a root-knot nematode infested field at the Plant Breeding Unit (PBU) located at the E.V. Smith Research Center of Auburn University near Tallassee, AL, in October of 2016. Galling was observed on the host plants in the field, confirming RKN presence (Fig. 1). The field has continually been used for root-knot nematode research for 10 years, and the crops planted in the field for the 2016 growing season were cotton, corn, and soybean, with these crops being in the same location for the previous two growing seasons. The field is a Kalamia loamy sand (80% sand, 10% silt, 10% clay). Three independent soil samples were taken based upon the crop planted in 2016. Soil samples were taken with a hand held conical soil probe (2.54 cm wide and 20 cm deep), and were taken evenly over the entire area of the field by walking a zigzag pattern over each area until a volume of 20-liter bucket was obtained per sample. Samples were mixed, and a 100-cm^3 soil subsample was processed to determine RKN population density. Nematodes were extracted by gravity sieving followed by sucrose centrifugation following the methodology of Barker (1985). After extraction, nematode presence was confirmed and enumerated via a Nikon TSX 100 inverted microscope at 40× magnification. This procedure was repeated three times for each soil sample, and the average RKN population counted was used for initial population density. Average RKN density was calculated at 72 second-stage juveniles (J2)/100 cm^3 of soil.

The soil from PBU was used for three different differential-host tests (DHT) identified by crop sampled. For the DHT, six host plants were planted in the RKN-infested soil and grown over a 45-day period to determine the host range and reproductive factor of the unknown RKN population. The test was modeled by Taylor and Sasser (1978), but modified to use juveniles vs. eggs as inoculum, and plant cultivars were

Figure 1. Symptomatic galling as a result of root-knot nematode infection on cotton roots.

substituted when the original cultivars could not be obtained. Field soil with RKN J2 of a known population density previously determined was used for the test. No other inoculation occurred. Host plants used in the test were cotton (Fibermax 1944), tobacco (NC 95), pepper (California Wonder), watermelon (Charleston Grey), peanut (Flor-runner), and tomato (Rutgers) (Sasser, 1972). For this test, corn (Mycogen 2R042; Dow AgroSciences, Indianapolis, IN) and soybean (Asgrow 5935; Monsanto Company, St. Louis, MO) were also included in the differential-host test. While these crops are not traditionally included in the DHT, they were included for reference as they were the original host of two of the three samples screened. The original study by Sasser (1972) used cotton cv. Deltapine 16. We were unsuccessful in finding an adequate supply of seed of this cotton cultivar, so Fibermax 1944 was used. FM 1944 has been shown to be susceptible to RKN and provides a suitable alternative to Deltapine 16 (Smith, 2015). Tomato and tobacco plants were germinated in potting soil and transplanted at a rate of two plants per pot. All other host plants were seeded directly in a 500- cm^3 polystyrene pot containing soil taken from PBU. Plants were thinned to two plants per pot immediately after seedling emergence. Each DHT was arranged in a randomized complete block design with five replications and repeated once. Plants were watered as needed to maintain soil moisture between 40 to 60% of the field capacity. Temperatures in the greenhouse ranged from 24 to 35°C with an average of 30°C, and lighting was supplied via 1000-watt halide bulbs producing 110,000 lumens at 14 hr/day.

The test was terminated 45 days after plant emergence, allowing for a complete life cycle of the RKN population to occur on the host plant. Plant shoots were cut from roots, and roots were gently washed under a faucet to remove loose soil. Root and shoot fresh weights were recorded, and RKN was extracted from 100 cm^3 soil. J2 were extracted from soil as previously described for calculating final density, and these numbers were then used to determine the reproductive factor (Rf) of each host. Reproductive factor was calculated by the following formula: Rf = final J2 population density at 45 days divided by initial J2 population density (72 J2/100 cm^3 of soil) as described by Oostenbrink (1966). The Rf values were then grouped into four categories and are as follows: Rf = 0-0.09, nonhost; Rf = 0.1-0.9, poor host; Rf = 1-2, moderate host; R > 2, suitable host (Oostenbrink, 1966). Rf values were statistically analyzed using ANOVA in SAS 9.4, and means were compared using Tukey-Kramer with $P \leq 0.05$.

All three samples. originally taken from field as populations and labelled AU76, AU77, and AU78, were identified as *M. incognita* race 3 through the use of the differential-host test (Table 1). The reproductive factor was highest on the crop that was the original host of the population in all three samples (Fig. 2). AU76 had an Rf = 8.7 on cotton, AU77 had a Rf = 4.1 on corn, and AU78 had a Rf = 6.6 on soybean. None of the three populations were a host of tobacco or peanut. All samples reproduced on cotton, pepper, watermelon, tomato, corn and soybean.

With all populations being identified as *M. incognita* race 3, the management implications for this field are the same despite coming from three different host crops. Whereas only one species was identified in this field, it is interesting that the Rf for each population was highest on the original host crop of the population — emphasizing an importance of crop rotation. AU76, coming from cotton, had an Rf = 8.7 on cotton, with AU 77 and AU 78, coming off corn and soybean, respectively, having Rf = values of 2.7 and 3.2 for cotton. AU77 had an Rf = 4.1 for corn, compared to AU76 at 1.7 and AU78 at 2.1. AU78 had an Rf = 6.6 on soybean, with AU76 and AU77 = 2.2 and 1.7, respectively. These numbers indicate that multiple populations of the same RKN species may respond in a slightly different manner if taken from different host crops. Similar variations have been seen for soybean cyst nematodes, where multiple HG types were reported in the same field but did

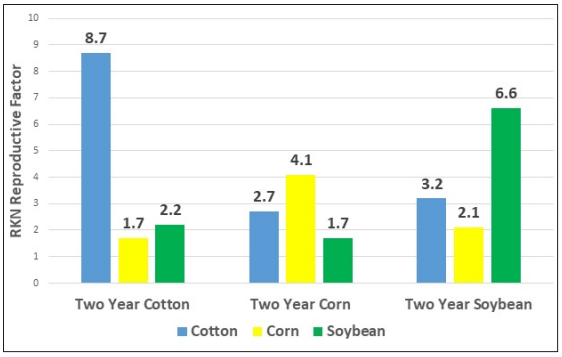

Figure 2: Reproductive factor of *Meloidogyne incognita* race 3 collected from a single field and grown on cotton, corn, and soybean in the greenhouse.

Table 1. Greenhouse differential-host test results for reproduction of *Meloidogyne* from three soil samples within a 1.5 ha area at the Auburn University Plant Breeding Unit in Tallassee, AL, across cotton, soybean, and corn crops with inoculum level at 72 second-stage juveniles/100 cm3 soil and final population densities at 45 days after plant emergence.

		Reproductive factor[x]								
Population	Original Host	Cotton	Tobacco	Pepper	Watermelon	Peanut	Tomato	Corn	Soybean	Species
AU76	Cotton	8.7 (+) a [yz]	0.2 (-) a	2.2 (+) a	1.8 (+) a	0.1 (-) a	3.5 (+) a	1.7 (+) b	2.2 (+) b	*M. incognita* race 3
AU77	Corn	2.7 (+) b	0.4 (-) a	1.9 (+) a	2.3 (+) a	0.0 (-) a	2.8 (+) a	4.1 (+) a	1.7 (+) b	*M. incognita* race 3
AU78	Soybean	3.2 (+) b	0.1 (-) a	3.4 (+) a	1.7 (+) a	0.0 (-) a	4.5 (+) a	2.1 (+) b	6.6 (+) a	*M. incognita* race 3

[x] Reproductive factor determined based upon methods described by Oostenbrink (1966).

[y] Column numbers followed by the same letter are not significantly different at $P \leq 0.05$ as determined by Tukey's multiple-range test.

[z] +/- designate determination of *Meloidogyne* population being a host or nonhost of the test plant.

not impact management (Beeman *et al*., 2016). However, data specific to this study show that RKN populations may have a host preference for the original crop they were sampled from.

Overall, this study shows the variability that can occur among a RKN population in a single field. RKN species differentiation is important and can impact a grower's crop rotation decision. While the RKN population is a parasite of all crops in this field, Rf calculations can predict how well the RKN population will reproduce on, and thus damage, each crop potentially grown in a field. Thus, when RKN population density is at damaging levels, implementing a crop rotation, even if to another host of the RKN population, can potentially limit damage by lowering the RKN reproductive factor.

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LITERATURE CITED

Barker, K. R. 1985. Nematode extraction and bioassays. Pp. 19-35 *in* Barker, K.R., C.C. Carter, and J.N. Sasser (eds.) An Advanced Treatise on *Meloidogyne:* Volume II. Methodology. Raleigh, NC, USA: North Carolina State University.

Beeman, A. Q., C. J. Harbach, C. C. Marett, and G. L. Tylka. 2016. Soybean cyst nematode HG type test results differ among multiple samples from the same field but management implications are the same. Plant Health Progress 17:160-162.

Davis, R. F., and P. Timper. 2000. Resistance in selected corn hybrids to *Meloidogyne arenaria* and *M. incognita.* Journal of Nematology 32:633-640.

Nicol, J., S. Turner, D. Coyne, L. den Nijs, S. Hockland, and Z. Tahna Maafi. 2011. Current nematode threats to world agriculture. Pp. 21-43 *in* Jones, J., and G. Godelieve (eds.) Genomics and Molecular Genetics of Plant-Nematode Interactions. London: Springer Science+Business Media B.V.

Oostenbrink, M. 1966. Major characteristics of the relation between nematodes and plants. Mededelingen Landbouhoge School, Wageningen 66:3-46.

Sasser, J. N. 1972. Physiological variation in the genus *Meloidogyne* as determined by differential hosts. OEPP/EPPO Bulletin 6:41-48.

Smith, A. 2015. Identification of resistant or tolerant commercial cotton cultivars to the Fusarium wilt root-knot nematode disease complex and the identification of *Fusarium oxysporum* f. sp. *vasinfectum* races in Alabama. Master's Thesis, Auburn University. https://etd.auburn.edu/handle/10415/4563.

Taylor, A. L. and J. N. Sasser. 1978. Biology, identification, and control of root-knot nematodes (*Meloidogyne* species). Raleigh, NC: North Carolina State University Graphics.

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