

RESEARCH/INVESTIGACIÓN

SEASONAL VARIATION AND COTTON-CORN ROTATION IN THE SPATIAL DISTRIBUTION OF *ROTYLENCHULUS RENIFORMIS* IN MISSISSIPPI COTTON SOILS

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ABSTRACT

Lee, H. K., G. W. Lawrence, J. L. DuBien, and K. S. Lawrence. 2015. Seasonal variation and cotton-corn rotation in the spatial distribution of *Rotylenchulus reniformis* in Mississippi cotton soils. *Nematropica* 45:72-81.

Horizontal and vertical distribution of the reniform nematode, *Rotylenchulus reniformis* Linford and Oliveira, was examined over a 3-yr period in Mississippi cotton soils. Two naturally infested cotton fields (8.1 ha each) were examined to determine the effects of continuous cotton production and a cotton-corn rotation system on the nematode population dynamics in two different seasons (spring and fall). Sixteen grids (0.4 ha) were established in each field. Both fields were grid-mapped with 16 points on 0.4-ha grids using a Global Positioning System (GPS). A single soil core, 5.08-cm diameter × 121.92 cm deep, was collected from each grid intersection and divided into 8 sample depths. Reniform nematodes were found at each of 16 geo-referenced sample sites and in all of the eight sampling depths. The number of reniform nematodes varied from 10 to 2,500 per 100 cm³ soil across sample sites. The nematode population appeared to have a non-clustered horizontal distribution across both the continuous cotton production and cotton-corn rotation fields. Reniform nematode numbers also differed by season. The sampling time and depth were associated with nematode density. In the continuous cotton production field, nematode numbers found in the upper soil profile were higher in the fall than in the spring. In the cotton-corn rotation field after corn was planted, more nematodes were recovered in the lower soil profiles in the fall. Corn and cotton rotations affected both nematode numbers and distribution in relation to soil depth. The recovery of reniform nematodes deep in the soil profile may help explain why this nematode is capable of reaching high levels in the soil after a single year rotation with corn.

Key words: cotton, cotton-corn rotation, horizontal distribution, population number, reniform nematode, *Rotylenchulus reniformis*, season, vertical distribution.

RESUMEN

Lee, H. K., G. W. Lawrence, J. L. DuBien, and K. S. Lawrence. 2015. Variación estacional y rotacional (algodón-maíz) en la distribución espacial de *Rotylenchulus reniformis* en suelos de algodones de Mississippi. *Nematropica* 45:72-81.

Se examinó la distribución horizontal y vertical del nemátodo reniforme, *Rotylenchulus reniformis* Linford and Oliveira, durante un periodo de 3 años en suelos de algodones en Mississippi. Se examinaron dos campos naturalmente infestados (8.1 ha cada) para determinar los efectos del monocultivo de algodón y de la rotación algodón-maíz sobre la dinámica de población del nemátodo en dos estaciones diferentes (primavera y otoño). Se establecieron dieciséis cuadrículas (0.4 ha) en cada campo. Ambas parcelas se cuadrícularon y mapearon con 16 puntos usando un sistema de posicionamiento global (GPS). Se recolectó una cata sencilla de suelo, 5.08-cm diámetro × 121.92 cm profundidad, en cada intersección de la cuadrícula, la cual se dividió en 8 submuestras por profundidades. Los nemátodos reniformes se encontraron en todas las muestras y en las 8 profundidades. El número de nemátodos reniformes varió entre 10 y 2,500 por 100 cm³ de suelo. La población del nemátodo parece tener una distribución horizontal no agrupada, tanto en el monocultivo de algodón como en los campos con rotación algodón-maíz. El número de nemátodos reniformes fue diferente según la estación. La fecha de muestreo y la profundidad de la muestra se asociaron a la densidad de nemátodos. En los campos con monocultivo

de algodón, el número de nemátodos recuperados de los perfiles superiores del suelo fueron mayores en otoño que en primavera. En los campos con rotación algodón-maíz, se encontraron más nemátodos en los perfiles inferiores del suelo en otoño. Las rotaciones de maíz y algodón afectaron tanto al número de nemátodos como a su distribución en relación a la profundidad del suelo. La recuperación de nemátodos reniformes desde las capas profundas del suelo puede explicar por qué este nemátodo es capaz de alcanzar niveles altos en suelo tras un año de rotación con maíz.

Palavras-chave: algodón, rotación algodón-maíz, distribución horizontal, densidad de población, nematodo reniforme, *Rotylenchulus reniformis*, estación, distribución vertical.

INTRODUCTION

Cotton (*Gossypium hirsutum* L.) is the most important textile fiber crop in the world. It is also the world's second most important oilseed crop after soybean (Lewis and Richmond, 1968; Poehlman and Sleper, 1995). In the United States, 17 states across the southern region are referred to as the Cotton Belt. In 2013, this region produced 13.2 million bales of cotton on 3.1 million ha. Mississippi is one of the highest cotton producing states based on the number of harvested acres (National Agricultural Statistics Service, 2014).

Four major plant-parasitic nematode species are known to parasitize cotton in the United States. These are the southern root-knot nematode, *Meloidogyne incognita* (Kofoid and White) Chitwood, the reniform nematode, *Rotylenchulus reniformis* (Linford & Oliveira), the Columbia lance nematode, *Hoplolaimus columbus* (Sher), and the sting nematode, *Belonolaimus longicaudatus* (Rau). Among these, the root-knot and reniform nematodes are considered the most serious nematode genera associated with cotton production (Starr, 1998). The more widespread of these two nematode pests is *M. incognita*, which has been widely distributed across the Cotton Belt for over 100 yrs. This nematode is found in all cotton-producing states in the United States. However, the reniform nematode is now recognized to have the potential to become a more serious pest on cotton. Recently, the reniform nematode has replaced the root-knot nematode as the greatest cause of nematode-induced economic losses and is considered to be the dominant parasitic species in cotton in the southeastern Cotton Belt (McLean and Lawrence, 2000; Overstreet and McGawley, 2002; Diez *et al.*, 2003). According to the Cotton Disease Loss Estimate Committee (Lawrence *et al.*, 2014), an average cotton yield loss due to diseases and nematodes was 12.6% in the United States in 2014. Yield losses caused by plant-parasitic nematodes alone were estimated at 4.99%. Subsequently, Mississippi's cotton yield losses due to nematodes averaged 8.6% or 57,600 bales annually

(Lawrence *et al.*, 2014).

The reniform nematode is found in 11 states including the Gulf Coast states, Arkansas, Georgia, North Carolina, and South Carolina (Heald and Robinson, 1990). Among these states, Alabama, Louisiana and Mississippi have the higher reniform incidence. The reniform nematode has been detected in 30%, 55%, and 32% of the cotton acreage of Alabama, Louisiana, and Mississippi, respectively (Lawrence and McLean, 1999). Cotton yield can be reduced 60% or more from heavy infestations of the reniform nematode in continuous cotton production. In Mississippi, the reniform is resident in 51 counties, and yield losses have been estimated at an average of 29% from infested fields (Lawrence and McLean, 2002).

Currently, there are no commercially available resistant cotton cultivars to manage the reniform nematode, although several cotton cultivars have displayed a degree of tolerance by producing acceptable yields when planted in fields infested with the reniform nematode (Robinson *et al.*, 1999; Koenning *et al.*, 2000). Nematicide application, crop rotation, and cotton cultivar selection are considered important practices in management of the reniform nematode (Lawrence and McLean, 1999, 2001, 2002). Corn, sorghum, peanut, wheat, and rice are nonhosts of the reniform nematode and can be effectively used as rotation crops. In Mississippi, corn is considered the best rotation crop for reniform nematodes (Gazaway, 1999; Lawrence and McLean, 2001, 2002; Overstreet and McGawley, 2002).

Most plant-parasitic nematodes that exist in agricultural fields feed on crop roots and may decrease yields. The degree of crop damage attributed to plant-parasitic nematodes depends on accurate measurements of nematode population density in the field. This knowledge is based on both the horizontal and vertical distribution of nematode populations (Ferris and McKenry, 1974; Van Gundy, 1985). The horizontal distribution of plant-parasitic nematodes is either uniform, random or clustered in an agronomic field (Starr, 1998). The most common pattern of horizontal distribution of nematodes is the

cluster (Norton, 1978). Unlike other plant-parasitic nematode species, the reniform nematode on cotton appears to have a non-clustered distribution throughout the field. Symptoms due to the reniform nematode are more or less uniformly distributed across the field, so unless a soil sample is collected, the nematode may not be detected. This unique type of distribution makes the reniform nematode difficult to detect based on damage to the cotton plant (Starr, 1998).

The vertical distribution of plant-parasitic nematodes changes over time as a result of different reproductive rates, mortality at particular depths from directional nematode migration induced by changes in environmental conditions, or the existence of host roots (Wallace, 1964; Forge *et al.*, 1998). When in the soil prior to parasitism, outside the host plant, most plant-parasitic nematodes appear to concentrate at a particular depth, which may vary with environmental factors in agricultural soils. Vertical distribution patterns are produced by orientation and movement to a particular soil zone where conditions are optimal for reproduction. This zone fluctuates with environment and season as temperature and moisture gradients change (Wallace, 1964).

In Alabama Moore *et al.*, 2010 determined that although irrigation had no significant effect on the movement of the reniform nematode, the male life stage moved faster from the point of infestation in soils that received water. Vermiform life stages of the reniform nematode were capable of moving horizontally 200 cm from an initial point of infestation. Studies on the reniform nematode spatial distribution are limited. Research on this ecological aspect of the nematode usually includes tests examining either the horizontal or the vertical distribution of the nematode. In Mississippi, preliminary studies on the horizontal and vertical distribution of the reniform nematode were reported by Lawrence *et al.*, 1994. They determined that the reniform nematode had a non-clustered spatial distribution over a 0.4-ha test plot and was detected to a depth of 45 cm. The study was conducted during the cotton-growing season and vertical movement of the nematode in the absence of the cotton host plant or the effects of the cooler winter soil temperatures could affect nematode movement differently. In a separate study in Mississippi, the reniform nematode was found at a depth of 120 cm in a cotton production field (Lee *et al.*, 2003). Robinson *et al.* (2005) examined the vertical distribution of the reniform nematode to a depth of 122 cm in 20 cotton fields across 6 cotton-producing states. They reported that the reniform nematode was distributed throughout the soil profile to a depth of 122 cm. Additional studies are needed to better understand the population distribution of the reniform nematode

and the effects of a non-host rotation crop on vertical movement of the nematode.

The objectives of our investigation were to: i) determine the horizontal and vertical distribution of the reniform nematode in a Mississippi cotton field; ii) observe seasonal effects on nematode population densities; and iii) examine the variation of nematode population numbers influenced by a cotton-corn rotation system.

MATERIALS AND METHODS

A 3-yr study was conducted to examine the horizontal and vertical distribution of the reniform nematode. Tests were conducted in cotton fields that were naturally infested with the reniform nematode and located near Glen Allan, MS. Two separate cotton fields, both 8.1 ha in size, were selected to study the effects of continuous cotton or a cotton-corn rotation on nematode population dynamics. The field in a continuous cotton production system (330 04' 19.88" N; 910 03' 21.70" W, elev 111 ft.) and the other field in a cotton-corn rotation system (330 04' 26.55" N; 910 03' 14.19" W, elev 106 ft.) were near each other. In the cotton-corn rotation field, corn was planted in year 1, and cotton followed in year 2 and year 3. Soil samples for nematode analysis were collected from both fields in the spring and fall for 3 yr.

Each field was arbitrarily divided into 16 georeferenced sites on 0.4-ha grids using a Global Positioning System (GPS). This enabled soil samples to be collected at the same site on each subsequent sampling date. A single soil core, 5.08 cm-d \times 121.92 cm deep, was collected from each grid intersection with a Model 4808 Concord Soil Sampler (Nietfeld Bodenprobe technik/Badbergen/North Germany). Each core was divided into eight samples by depth: 0-15 cm, 16-30 cm, 31-45 cm, 46-60 cm, 61-75 cm, 76-90 cm, 91-105 cm, and 106-120 cm. Core samples were placed in plastic bags and labeled with location of the core site and sample depth. Soil cores were collected from the continuous cotton production field in June of year 1, May and November of year 2, and March of year 3. Soil cores were collected from the cotton-corn rotation field in September of year 1, May and November of year 2, and March of year 3.

Nematodes were extracted from each section of the soil core using gravity screening and centrifugal flotation (Jenkins, 1964), and reniform nematodes were counted with a stereo-microscope. The experiment was conducted in both continuous cotton and cotton-corn rotation production systems using a 4 \times 8 factorial arrangement of collection times and sampling depths replicated across 16 sites in each field. The data for the two fields were pooled for the three common sampling times in years 2 and 3, and analyzed in a three-way

cross classification analysis of variance (ANOVA) with factors of field, time, and depth on mean reniform nematode numbers using PROC GLM from SAS.

RESULTS

Reniform nematodes were found at each of 16 sample sites and eight sample depths in the field planted to continuous cotton (Fig. 1). In general, the reniform nematode appeared to have a relatively even horizontal distribution across both fields. However, the numbers of the reniform nematodes varied at each sample site and depth.

The field \times time \times depth interaction was significant at the 0.05 level with a *P*-value of 0.027 (Table 1), so the effects of collection time and sampling depth on reniform nematode population densities are not the same in the continuous cotton production field as in the cotton-corn rotation field.

In the continuous cotton production field, reniform nematodes were found in most of the 16 collection sites and eight depths in spring of year 1, spring and fall of year 2, and spring of year 3. Reniform numbers differed across sample sites

and the eight soil depths. Nematodes were found horizontally and vertically throughout the field. The response surface for reniform nematode numbers as a function of depth and time illustrates how reniform numbers changed by season within the soil depths (Fig. 2). The main nematode population change occurred between 45-cm and 90-cm depths through the three seasons. In the fall of year 2, the highest number of nematodes was observed (Fig. 2). The average number of nematodes for the 16 sites in the continuous cotton field in the spring (year 1) ranged from 374 to 1,428 nematodes per 100 cm³ soil. In year 2, averages in the spring ranged from 315 to 2,004. Population densities in the fall ranged from 199 to 788 and 101 to 839 nematodes per 100 cm³ for the fall of years 2 and 3, respectively (Fig. 3).

In the cotton-corn rotation field, soil samples were recovered at all 16 sample sites in fall of year 1 after corn, in both the spring and fall of year 2 during the cotton growing season, and spring of year 3 before cotton planting. Reniform nematodes were found in 77% of the 128 collection sites from the 8 soil depths. This is in contrast with the continuous cotton production field where we found only a very small

Table 1. A three-way cross classification ANOVA with factors of field, time, and depth in a continuous cotton production and cotton-corn rotation system^w.

Dependent Variable:	The GLM Procedure					
	Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
<i>Rotylenchulus reniformis</i> density						
Model	47	105859458.9	2252328.9	4.34	<.0001	
Error	720	373980553.3	519417.4			
Corrected Total	767	479840012.2				
<i>Rotylenchulus reniformis</i> density		R-Square	Coeff Var	Root MSE	Means	
		0.22061	122.3677	720.7062	588.967	
Source	DF	Sum of Squares	Mean Square	F Value	Pr > F	
Field ^x	1	770196.67	770196.67	1.48	0.2237	
Time ^y	2	8058287.89	4029143.94	7.76	0.0005	
Field*Time	2	33297844.34	16648922.17	32.05	<.0001	
Depth ^z	7	28238709.32	4034101.33	7.77	<.0001	
Field*Depth	7	5182937.13	740419.59	1.43	0.1918	
Time*Depth	14	16748549.89	1196324.99	2.3	0.0043	
Field*Time*Depth	14	13562933.64	968780.97	1.87	0.027	

^wThe 16 sampling sites served as replicates in each field.

^xField : continuous cotton production and cotton-corn rotation systems.

^yTime: spring year 2, fall year 2 and spring year 3.

^zDepth: in 15 cm increments from 0 to 120.

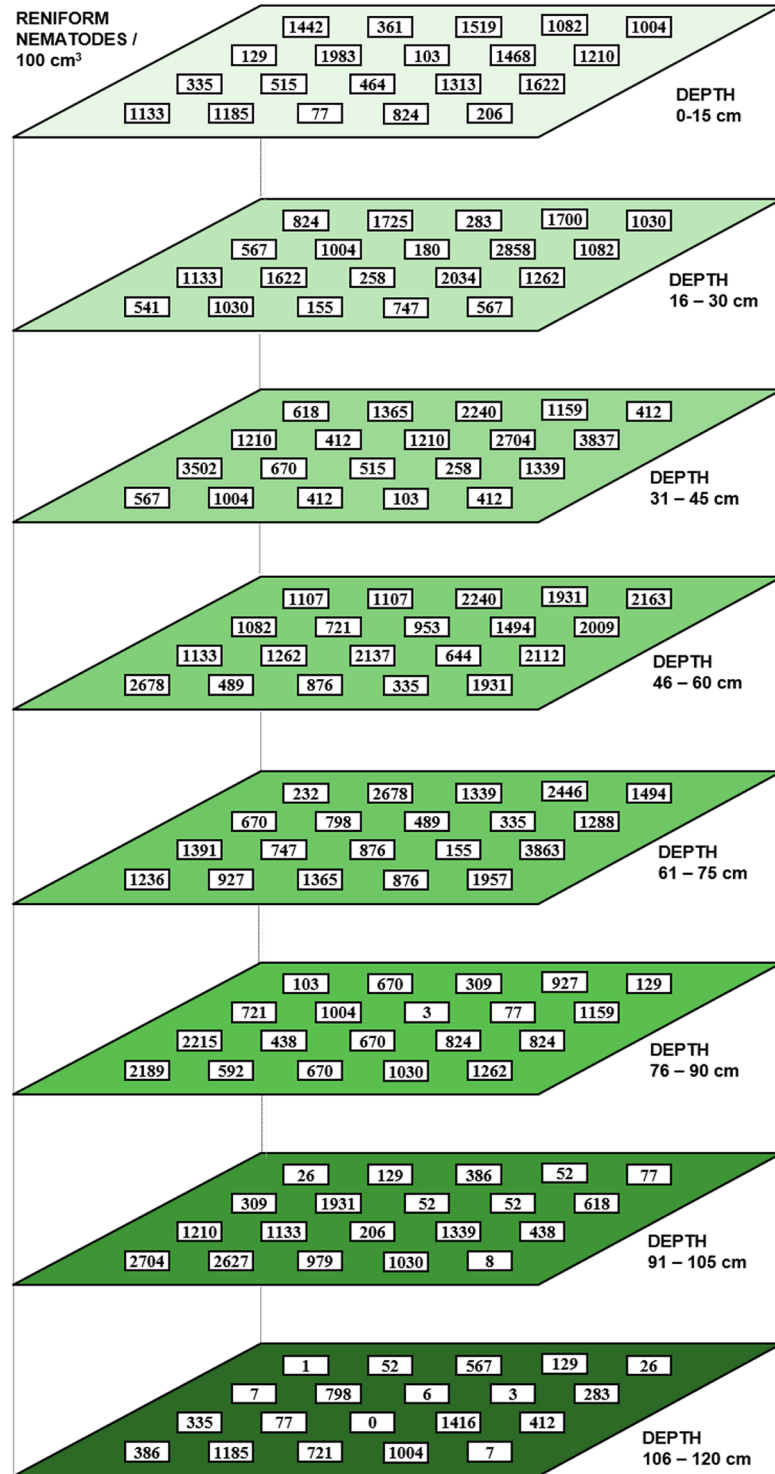


Fig. 1. The horizontal and vertical distribution of *Rotylenchulus reniformis* in a Mississippi field continuously planted with cotton. Each sample location represents the average number of *R. reniformis* juvenile and vermiform adult life stages/100 cm³ soil from the two field locations used in the study.

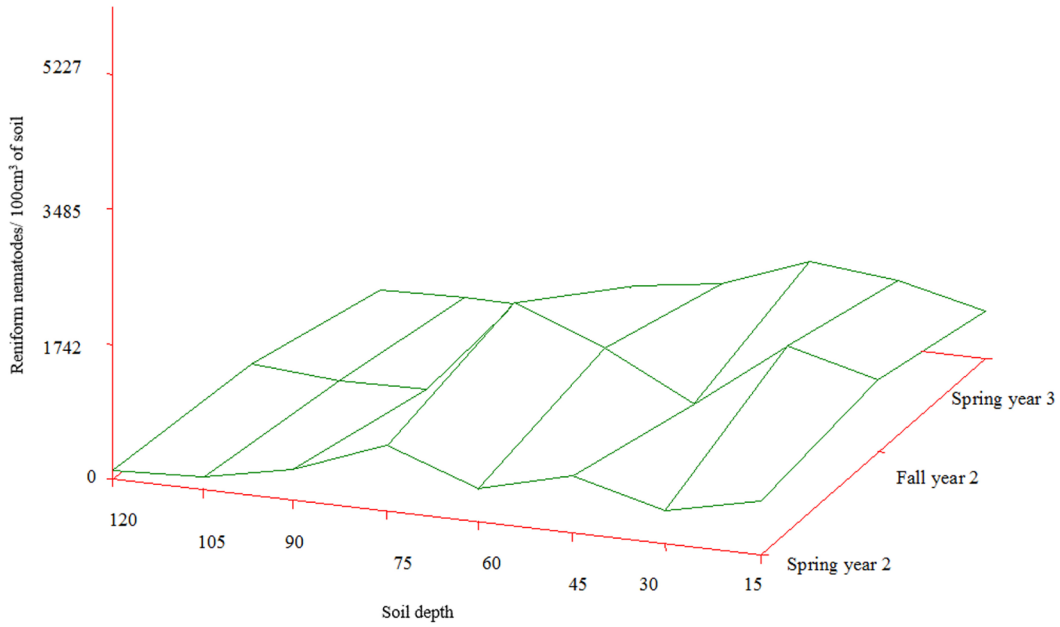


Fig. 2. Three-dimensional projection of seasonal change in *Rotylenchulus reniformis* population numbers at 8 soil depths in the continuous cotton production planting system. Each point represents the average of 16 sampling sites for each soil depth.

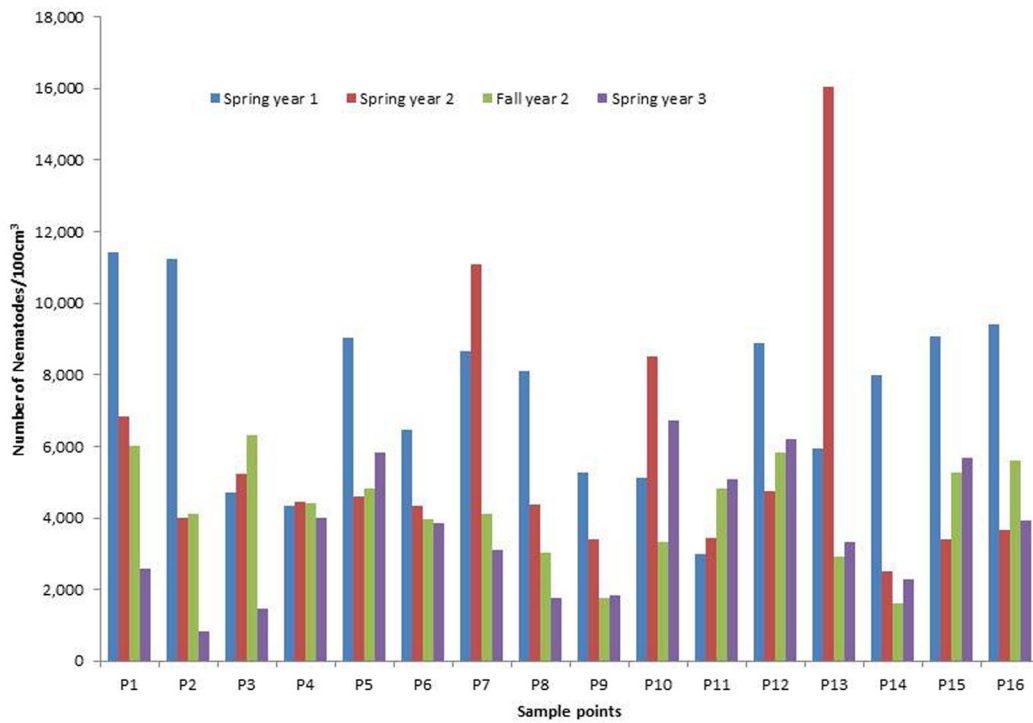


Fig. 3. Horizontal distribution of *Rotylenchulus reniformis* in the continuous cotton production system. Each value represents the total number of nematodes recovered at each sample point for all sample depths.

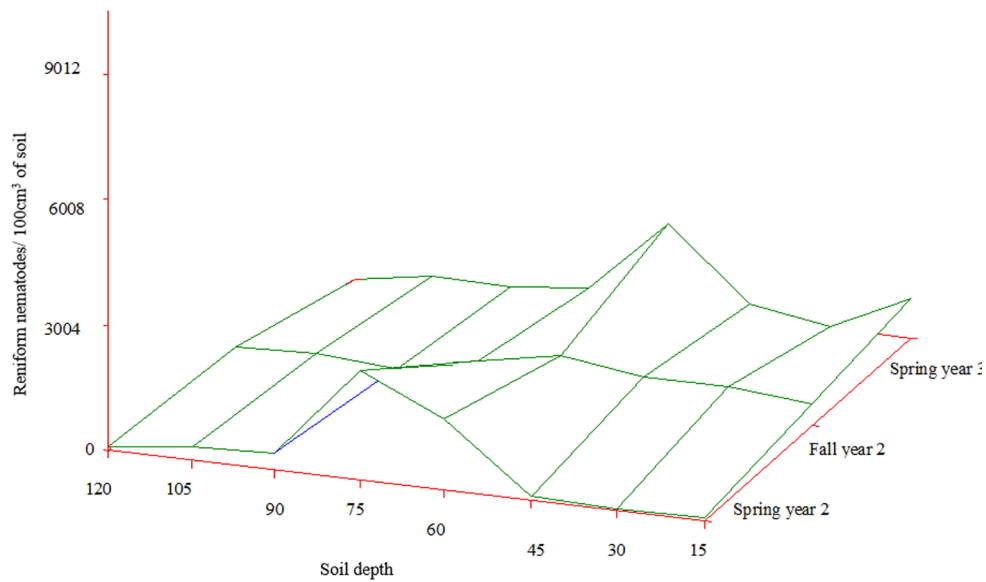


Fig. 4. Three-dimensional projection of seasonal change in *Rotylenchulus reniformis* population numbers at 8 soil depths in the cotton-corn rotation production planting system. Each point represents the average of 16 sampling sites for each soil depth.

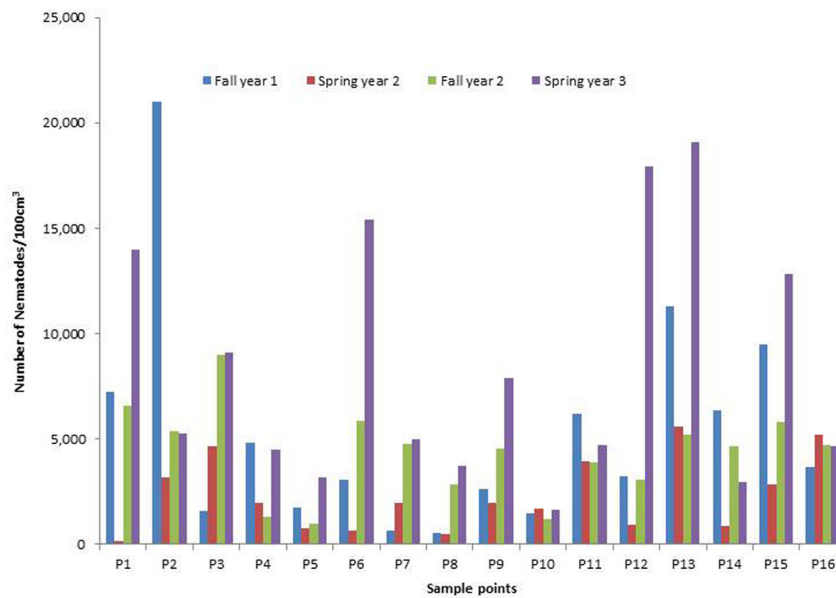


Fig. 5. Horizontal distribution of *Rotylenchulus reniformis* in the cotton-corn production system. Each value represents the total number of nematodes recovered at each sample point for all sample depths.

percentage of sites with no nematodes. Reniform nematode numbers differed across sample site and depth, and nematodes were found horizontally and vertically throughout the field.

Overall, the reniform nematode numbers were lower during the cotton growing season in spring and fall of year 2 after corn had been planted in year 1. By the spring of year 3, nematode numbers had increased after planting cotton for a single year. From the response surface of reniform nematode numbers as a function of time and depth, it can be observed that the main changes in nematode population occurred between 45 cm and 90 cm depths through the three seasons tested (Fig. 4). Interestingly, in the fall of year 2, the lowest numbers of nematodes were observed.

The average number of nematodes for the 16 sample sites ranged from 70 to 2,623 nematodes per 100 cm³ soil after corn in fall of year 1. The average number of nematodes for the 16 sampling sites ranged from 22 to 701 nematodes in spring of year 2; 122 to 1,126 nematodes in fall of year 2; and 206 to 2,388 nematodes in spring of year 3 when cotton was planted in the field (Fig. 5).

DISCUSSION

Unlike other plant-parasitic nematodes, the reniform nematode has a unique, non-clustered distribution pattern in a cotton field (Lawrence *et al.*, 1994; Lee *et al.*, 2003). The nematode does not seem to be limited by soil type, soil texture, soil pH or the amount of rainfall. However, finer-textured silt soils seem to support nematode populations more readily than coarser-textured, sandy soil (Heald and Robinson, 1990; Koenning *et al.*, 1996; Herring *et al.*, 2010). In our study, the reniform nematode had a non-clustered horizontal distribution across both the continuous cotton and cotton-corn rotation fields. Reniform nematodes were found in all of the 16 sample sites although the densities differed across sites and for each sampling time.

A non-clustered distribution was also reported by Moore *et al.* (2010), who suggested that once introduced into a field, reniform nematodes were capable of rapid horizontal movement. In their study, the nematode was able to reach a density that was damaging to cotton in 2 yr.

The reniform nematode is also capable of surviving deep in the soil profile. This is generally limited to the depth of root penetration by the host plant (Robinson *et al.*, 2005), although other environmental factors may influence vertical nematode movement including heat or other biological antagonists (Robinson *et al.*, 2005). This is an important consideration when estimating

nematode numbers for choosing and implementing management practices (Robinson *et al.*, 2005). In Tennessee, Newman and Stebbins (2002) found the reniform nematode to a depth of 90 cm in the soil. The highest numbers of nematodes were found at the 61 to 75 cm depth at planting. The population density gradually increased in the upper 46 cm of soil from midseason to harvest and the greatest population fluctuations were detected in the upper 30 cm. In our study we found that the reniform nematode was able to exist at a depth of at least 120 cm, which was the maximum depth of our equipment. Reniform nematodes were present at all depths. The greatest influence of time on nematode population densities was observed at a depth of 76 to 90 cm.

Reniform nematode numbers were higher in the fall than in the spring in the continuous cotton production system as would be expected for a susceptible, annual crop. Higher nematode numbers were found in the upper soil profiles in the fall compared to samples collected in the spring, and numbers decreased with increasing soil depth.

Rotation to corn for 1 yr lowered reniform nematode numbers. After 1 yr in corn, more nematodes were recovered in the lower soil profiles in the fall. The recovery of the reniform nematode deeper in the profile after a single year in corn may help explain why this nematode is capable of reaching high levels after a single year in cotton following the rotation crop. Gazaway *et al.* (1998) reported reniform nematode numbers that were lowered in a 1-yr and 3-yr rotation rebounded to damaging levels after one season of cotton.

Although, historically, crop rotation was not an option for many cotton producers because of the absence of an economically viable alternative to cotton (Overstreet and McGawley, 2002; Zimet *et al.*, 2002), in Mississippi corn is now considered the best rotation crop where the reniform nematode is the primary nematode pest. Unfortunately, however, corn is a host of the root-knot nematode. Fields infested with both root-knot and reniform nematodes may not benefit as much from a corn rotation because corn will allow the root-knot nematode population to increase. Where reniform occurs without the presence of root-knot, both its horizontal and vertical distribution are important when devising and implementing a reniform nematode management plan.

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