WEED HOSTS AND RELATIVE WEED AND COVER CROP SUSCEPTIBILITY TO *ROTYLENCHULUS RENIFORMIS* IN THE MISSISSIPPI DELTA

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ABSTRACT

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The reniform nematode (*Rotylenchulus reniformis*) causes economic losses in cotton and soybean in the southeastern United States, and has the ability to reproduce on more than 300 plant species. Even when the host crop is protected through the use of nematicides or host plant resistance, the potential exists for other plants present in or near the field to support the reniform nematode population. Because of variations in weed susceptibility, nematode virulence, and environmental factors, determining the host status of weeds to local populations of the nematode is needed to provide management recommendations. To identify the most important weed and cover crop hosts for reniform nematodes in the Mississippi Delta, nematode infection was measured on 53 plant species in greenhouse and field surveys. Subsequent greenhouse and field tests were conducted to confirm the host status of nine of these plants. Sicklepod (*Senna obtusifolia*), spurred anoda (*Anoda cristata*), entireleaf morningglory (*Ipomoea* sp.), and velvetleaf (*Abutilon theophrasti*) were identified as excellent hosts for reniform nematode in this region, supporting nematode populations equivalent to or greater than those developing on susceptible cotton plants included in the tests. Purple (*Cyperus rotundus*) and yellow (*C. esculentus*) nutsedges were poor hosts for reniform nematode, despite all underground plant parts supporting nematode infection. Sicklepod, velvetleaf, and entireleaf morningglory were reported to be important hosts for reniform nematode in tests in Alabama and Georgia indicating that efforts to manage these weeds across a wider geographic region may be needed.

Key words: hosts, nematode, reniform, *Rotylenchulus*, weeds.

RESUMEN

Molin, W. T., y S. R. Stetina. 2016. Malas hierbas hospedadoras y susceptibilidad relativa de malezas y cultivos de cobertura a *Rotylenchulus reniformis* en el delta del Misisipi. Nematropica 46:121-131.

El nematodo reniforme, *Rotylenchulus reniformis* causa pérdidas económicas en los cultivos de algodón y soja del sudeste de los Estados Unidos de América, y tiene la capacidad de reproducirse en más de 300 especies de plantas. Incluso cuando el cultivo hospedador está protegido a través del uso de nematicidas o de resistencia vegetal, el potencial de la población de nematodos reniformes para mantenerse en otras plantas presentes en el propio campo o parcelas cercanas existe. Debido a la variabilidad en la susceptibilidad de las malezas, virulencia de las poblaciones del nematodo y factores ambientales, se necesita determinar el estatus como hospedador de las malezas a las poblaciones locales del nematodo, con el objetivo de proporcionar recomendaciones para su manejo. Para identificar las malezas y cultivos de cobertura más importantes del delta del Misisipi, se midió la infección por el nematodo en 53 especies de plantas en invernadero y prospecciones en campo. Se realizaron ensayos adicionales en invernadero y campo para confirmar el estatus hospedador de nueve de estas plantas. EL senna chino (*Senna obtusifolia*), malva (*Anoda cristata*), campanillas (*Ipomoea* sp.) y abutilón (*Abutilon theophrasti*) fueron identificadas como excelentes hospedadores del nematodo reniforme en la región, manteniendo poblaciones equivalentes o mayores que aquellas que se desarrollaron en plantas de algodón susceptible que se incluyeron en los ensayos. Los coquillos púrpura (*Cyperus rotundus*) y amarillo (*C. esculentus*) fueron hospedantes pobres para el nematodo reniforme, a pesar de que todas las partes subterráneas de las planta mantenían la infección por nematodos. Senna chino, abutilón y campanillas, han sido citados como hospedadores importantes para el nematodo reniforme en ensayos en Alabama y Georgia indicando que los esfuerzos para el manejo de estas malezas pueden ser necesarios a un nivel geográfico más amplio.

Palabras clave: hospedadores, nematodo, reniforme, *Rotylenchulus*, malezas.

INTRODUCTION

The reniform nematode (*Rotylenchulus reniformis* Linford & Oliveira) is widely distributed throughout the southeastern and midsouthern sections of the United States. Two major crops grown in this region, cotton (*Gossypium hirsutum* L.) and soybean (*Glycine max* L.), are hosts and suffer economic losses when infected by this nematode (Kinloch, 1980; Robinson, 2007). The nematode has a broad host range including more than 300 plant species (Robinson *et al*., 1997; Khan, 2005), so even when these host crops are protected through the use of nematicides or host plant resistance (Kinloch, 1980; Starr *et al*., 2007), the potential exists for other plant species present in or near production fields to support the reniform nematode population. While weeds are the most likely noncrop hosts, cover crops, or volunteer plants from a prior season's crop could also serve to support reniform nematode reproduction.

Knowing which species in the local production area have the greatest potential to serve as hosts for the reniform nematode is important so they can be selectively targeted. Reports of variability among reniform nematode populations at the genomic level (Agudelo *et al*., 2005; Arias *et al*., 2009; Leach *et al*., 2012) and with respect to reproduction and pathogenicity (McGawley *et al*., 2010, 2011) underscore the importance of assessing the host status of weeds to local populations of the nematode to perhaps provide better recommendations for management in the region.

Some of the more important weeds in cotton and soybean production in the southeastern U.S. that are also hosts for reniform nematode include morningglory species, pigweed species, sicklepod, and prickly sida (Davis and Webster, 2005). Not only are these weeds among the most prevalent and troublesome in cotton (Webster and Nichols, 2012; Webster, 2013), but they have been projected to be among the most troublesome in reduced tillage systems (Bryson and Keeley, 1992), a current row crop production strategy widely used in the southeastern U.S. Two members of the Cyperaceae, yellow (*Cyperus esculentus*) and purple nutsedge (*C. rotundus*), which are also important weeds in cotton and soybean, have been reported both as hosts and nonhosts. Reniform nematode egg production in purple nutsedge considered as a percentage relative to egg production in cotton was 12.9% (943 eggs) and 453.5% (114,686 eggs) in two trials, whereas in yellow nutsedge egg production was zero and 10.9% (2743 eggs) (Davis and Webster, 2005). These values were higher than those for several broadleaf weeds known to support reniform nematode populations. In contrast, very low levels of infection were reported for yellow and purple nutsedge compared with cotton (Lawrence *et al*., 2008) and compared with infection rates of other weeds (Quénéhervé *et al*., 2006). These results indicate that the level of nematode infection on weedy hosts may vary by location or method of measuring infectivity, and may provide conflicting results. Considering the variability in response of sedges to infection, further examination of sedge responses to reniform nematode are warranted.

A common procedure for evaluating weed susceptibility to reniform nematode is to challenge seedlings by inoculating them with eggs harvested from a susceptible crop, waiting for 2 mo, and then harvesting the roots and quantifying the newly-formed eggs (Davis and Webster, 2005; Lawrence *et al*., 2008). A better approach may be to grow weeds side by side with a crop and examine roots of both plants for infection by reniform nematode under field conditions. Pontif and McGawley (2007) evaluated reniform nematode reproduction on cotton grown alone or in close proximity to morningglory (*Ipomoea lacunosa*), hemp sesbania (*Sesbania exaltata*), or johnsongrass (*Sorghum halepense*) and found that nematode reproduction on cotton was reduced when the crop was co-cultured with weeds. Complimentary work by these researchers demonstrated that weed root leachates inhibited reniform nematode reproduction on cotton, suggesting that allelochemicals could be involved in the response.

In addition to possible variability in nematode virulence and biotype, weed susceptibility and ability to sustain production of new egg masses, and differences in environmental influences between locations may affect host status determination at a given location. The objectives of this research were to survey weeds common in Mississippi cotton fields to determine their potential importance as hosts for reniform nematode, perform an in-depth examination of the potential sites of infection in yellow and purple nutsedge to more clearly define the role that sedges play in sustaining reniform nematode populations in the field, and confirm the host status of select weeds in standard greenhouse tests as well as under field conditions.

MATERIALS AND METHODS

Greenhouse weed and cover crop survey

To identify the most important weed and cover crop hosts for reniform nematode in the Mississippi Delta, 53 plant species (51 weeds and two cover crops) were initially screened in greenhouse surveys. These plants represented 14 botanical families:

Amaranthaceae (2 entries), Asteraceae (6 entries), Brassicaceae (2 entries), Caryophyllaceae (2 entries), Chenopodiaceae (1 entry), Convolvulaceae (5 entries), Cyperaceae (2 entries), Fabaceae (7 entries), Linaceae (1 entry), Malvaceae (3 entries), Poaceae (17 entries), Polygonaceae (2 entries), Portulacaeae (1 entry), and Solanaceae (2 entries). Due to greenhouse space constraints, plant species were divided into four different sets (tests 1 through 4) for evaluation. Species examined in each test are listed in Tables 1 through 4, and each test included one or more species that had previously been reported to serve as a host for reniform nematode.

Small clay pots were filled with 200 cm^3 of steam-sterilized soil mix (3 parts sandy loam soil: 2 parts sand). Four pots were sown with seed of each weed or cover crop species. Approximately 2 weeks later, plants were thinned to a final density of 4 plants per pot. After thinning, each pot was inoculated with 10,000 reniform nematodes (mixed vermiform stages) suspended in 3 ml water. Three holes, each approximately 2-cm deep, were made in the soil, and the nematode suspension was pipetted equally into the holes. Reniform nematode isolate RR02, originally derived from a single egg mass collected from a cotton plant in Elizabeth, MS in 2003, was used for all tests. This isolate was maintained on tomato (*Solanum lycopersicum* L. 'Rutgers') plants in a greenhouse. After inoculation, pots were arranged in a completely randomized design on greenhouse benches.

Four weeks after inoculation, plant roots were separated from soil by soaking the pot in water and gently agitating the root ball while submerged to remove any remaining soil mix. The plants from each pot were separated from each other, and the root system was removed by cutting at the soil line. Roots were stained with red food coloring following published protocols (Thies *et al*., 2002), and female nematodes infecting the roots were counted at ×30 magnification. After counting, the root system was placed briefly on a paper towel to drain excess liquid, and each root system was weighed. Counts from individual root systems were expressed as the number of females per gram of root. Each experiment was performed once.

Field weed survey

On 26 June 2015, naturally-occurring weeds in a cotton research field located near Stoneville, MS were collected and examined to determine if they were infected by reniform nematode. Plants were collected from multiple locations within the field for each of the following species: sicklepod (10 plants from 10 locations), prostrate spurge (*Euphorbia maculata* L.; 30 plants from 16 locations), hemp sesbania (10 plants from 8 locations), velvetleaf (10 plants from 10 locations), purslane (11 plants from 9 locations), Palmer amaranth (14 plants from 10 locations), and teaweed (15 plants from 13 locations). Plants were dug from the field using a shovel such that a soil ball approximately 20 cm in diameter surrounding the plant was removed. Roots were knocked free of surrounding soil, placed in a plastic bag, and returned to the lab for processing using the root-staining protocol previously described. Nematode counts were expressed as number of females (swelling, swollen, and gravid stages combined) per gram of root. A 200-cm^3 subsample of the soil surrounding the roots was processed using standard elutriation (Byrd *et al*., 1976) and sucrose centrifugation (Jenkins, 1964) procedures to confirm the presence of reniform nematode in the vicinity of each plant. Reniform nematodes were counted at ×50 magnification.

Host status confirmation in greenhouse tests

Reniform nematode population development on nine weed species that were demonstrated to support relatively high levels of infection by reniform nematode in the greenhouse survey was compared to that on susceptible cotton in a greenhouse experiment. Seeds of common waterhemp, entireleaf morningglory, hemp sesbania, northern jointvetch, sicklepod, spurred anoda, teaweed, velvetleaf, yellow sweetclover, susceptible cotton cv 'Stoneville 4892 BG/RR' (Bayer CropScience, Research Triangle Park, NC, USA), resistant *Gossypium arboreum* accession A2-190 (PI 615699), and resistant corn hybrid 'Pioneer 3223' (Pioneer Hi-Bred International, Johnston, IA, USA) were planted in clay pots containing 1,360 cm^3 of the same soil mix used in the greenhouse survey. After stand establishment, plants were thinned to one plant per pot and inoculated with 5,000 reniform nematodes (mixed vermiform stages) in 3 ml water using the same isolate and inoculation method as described for the greenhouse survey. After inoculation, five pots of each plant species were arranged in a completely randomized design on the greenhouse benches.

Eight weeks after inoculation, the test was harvested. Six soil cores 2.5-cm in diameter, representing the entire depth of soil in each pot, were collected and combined. Nematodes were extracted from a 100-cm^3 subsample of the soil in each pot using elutriation and sucrose centrifugation and counted as previously described. The tops of the plants were removed by cutting at the soil line and discarded. Plant roots were separated from

remaining soil and weighed. Roots were cut into pieces approximately 2.5-cm in length, and a random subsample of pieces with a final combined weight of 3 g was collected. Eggs were extracted from this root sample by stirring for 10 min in a 0.6% NaOCl solution (Hussey and Barker, 1973) and collecting eggs on a standard 25-µm-pore sieve. Nematodes and eggs were counted at ×50 magnification. The experiment was repeated and data from both trials were combined for analysis.

Nutsedge field survey

Purple and yellow nutsedge plants were collected on 26 June 2015 from borders of the same cotton field used for the field weed survey. A total of 10 plants of each species was collected. Care was taken to harvest all roots, rhizomes, and tubers as well as the soil surrounding the plants to a distance of 8 cm from the center of the plant. Rhizome, tuber, and root tissues from each plant were separated, then stained with red food coloring and weighed as previously described. The number of female reniform nematodes (swelling plus swollen) infecting each tissue type was determined at ×30 magnification, and counts were expressed as the number of females per gram of the respective plant tissue. To confirm that reniform nematodes were present in the soil adjacent to the plants, nematodes were extracted from a 200-cm^3 subsample of the soil surrounding each nutsedge plant using elutriation and sucrose centrifugation as previously described. Reniform nematodes were counted at ×50 magnification.

Host status confirmation in field tests

Two field locations with soils naturally infested with reniform nematodes were identified near Stoneville, MS. Both field locations were planted with the same susceptible cotton cultivar (ST 4946GLB2, Bayer CropScience, Research Triangle Park, NC, USA) on 3 May 2015. The host status confirmation test was established at each location on 28 May 2015. Plant species evaluated in this study included five weed species identified as potentially important hosts in greenhouse tests: sicklepod, spurred anoda, velvetleaf, entireleaf morningglory, and teaweed. Each plant species was replicated 4 times at each location. Twenty seed of each plant species were placed in an 8-cm long furrow 1-cm deep and 10 cm from the existing cotton row. A cottonseed of cultivar 'ST 4946GLB2' was planted at each end of the furrow so that cotton and test plants would emerge together. The micro-rows were hand irrigated as needed.

On 26 June 2015, the sown weeds and adjacent cotton plants were collected and examined to determine if they were infected by reniform nematode. Plants were dug from the field using a shovel such that a soil ball approximately 20 cm in diameter surrounding the plants was removed. Roots were knocked free of surrounding soil, placed in a plastic bag, and returned to the lab. Cotton and weed roots were carefully separated from each other prior to processing using the root staining protocol previously described. To be sure sufficient tissue was available for infection by the nematodes, data were not collected on root systems shorter than 5 cm. The number of swelling plus swollen females infecting the roots of each plant was expressed per gram of root tissue. A 200-cm^3 subsample of the surrounding soil was processed using the elutriation and sucrose centrifugation protocols previously described to confirm that reniform nematode was present in each micro-row, and the cotton roots were examined to confirm that reniform nematodes were active and infecting plants during the test period. Data from both locations were combined for analysis.

Statistical analysis

To normalize data, nematode counts were transformed [$\log_{10}$ (x + 1)] prior to analysis of variance (ANOVA). Backtransformed (geometric) means are presented. Where ANOVA showed significant differences in nematode numbers, means separation was determined based on differences of least squares means ($P \leq 0.05$). All analyses used SAS statistical software (PROC MIXED of SAS version 9.3; SAS Institute, Cary, NC, USA). For host status confirmation tests that were repeated in time (greenhouse) or space (field), initial data analyses (data not shown) identified no significant differences between trials, and no significant interactions between trial and plant species with respect to reniform nematode populations in soil or roots. Therefore, data from both trials were combined for final analysis, and trials and their interactions were modeled as random effects.

RESULTS

The numbers of reniform nematode females developing on plants in the survey conducted in the greenhouse are summarized in Tables 1 through 4. During the test period, most of the nematodes attached to the roots had developed to the fully swollen or gravid stages of development. Common groundsel and white clover supported the most nematodes of any of the plants surveyed (Table 3). Additional weed species that supported relatively high levels of reniform nematode infection were hemp sesbania

(Table 1), teaweed (Table 1), sicklepod (Table 1), entireleaf morningglory (Table 2), sowthistle (Table 2), common chickweed (Table 2), velvetleaf (Table 2), spurred anoda (Table 2), yellow sweetclover (Table 2), mouseear chickweed (Table 3), northern jointvetch (Table 3), and common waterhemp (Table 4). Nematodes were not found on roots of the cover crops common flax and cereal rye or on the roots of 12 weed species surveyed: barnyardgrass, browntop millet, crabgrass, Italian ryegrass, johnsongrass,

Table 1. Number of reniform nematode females developing on ten weed and two cover crop[y] species surveyed in greenhouse test 1.

Common name of plant	Scientific name of plant	Females per g root[z]
hemp sesbania	*Sesbania herbacea* (P. Mill.) McVaugh	49 a
teaweed	*Sida spinosa* L.	30 b
sicklepod	*Senna obtusifolia* (L.) H.S. Irwin & Barneby	18 c
pitted morningglory	*Ipomoea lacunosa* L.	11 d
barnyardgrass	*Echinochloa crus-galli* (L.) Beauv.	0 e
browntop millet	*Urochloa ramosa* (L.) Nguyen	0 e
crabgrass	*Digitaria* sp.	0 e
common flax[y]	*Linum usitatissimum* L.	0 e
Italian ryegrass	*Lolium perenne* L. ssp. *multiflorum* (Lam.) Husnot	0 e
johnsongrass	*Sorghum halepense* (L.) Pers.	0 e
cereal rye[y]	*Secale cereale* L.	0 e
yellow nutsedge	*Cyperus esculentus* L.	0 e
F		93.79
P > *F*		< 0.0001

[z]Values are means of up to 16 replications; means followed by the same letter are not significantly different based on differences of least squares means ($P \leq 0.05$).

Table 2. Number of reniform nematode females developing on 14 weed species surveyed in greenhouse test 2.

Common name of plant	Scientific name of plant	Females per g root[z]
entireleaf morningglory	*Ipomoea* sp.	102 a
sowthistle	*Sonchus* sp.	75 ab
common chickweed	*Stellaria media* (L.) Vill.	30 abc
velvetleaf	*Abutilon theophrasti* Medik.	36 abc
spurred anoda	*Anoda cristata* (L.) Schlecht.	18 abc
yellow sweetclover	*Melilotus officinalis* (L.) Lam.	14 bc
shepherd's-purse	*Capsella bursa-pastoris* (L.) Medik.	11 c
sprangletop	*Leptochloa* sp.	8 c
wild mustard	*Sinapis arvensis* L.	8 c
horseweed	*Conyza canadensis* (L.) Cronq.	7 cd
eastern black nightshade	*Solanum ptychanthum* Dunal	7 cd
Palmer amaranth	*Amaranthus palmeri* S. Wats.	1 de
junglerice	*Echinochloa colona* (L.) Link	0 e
purple nutsedge	*Cyperus rotundus* L.	0 e
F		5.83
P > *F*		< 0.0001

[z]Values are means of up to 16 replications; means followed by the same letter are not significantly different based on differences of least squares means ($P \leq 0.05$).

Table 3. Number of reniform nematode females developing on 20 weed species surveyed in greenhouse test 3.

Common name of weed	Scientific name of weed	Females per g root[z]
common groundsel	*Senecio vulgaris* L.	299 a
white clover	*Trifolium repens* L.	238 ab
mouseear chickweed	*Cerastium fontanum* ssp. *vulgare* (Hartman) Greuter & Burdet	36 bc
northern jointvetch	*Aeschynomene virginica* (L.) B.S.P.	36 bc
coffee senna	*Senna occidentalis* (L.) Link	9 cd
purslane	*Portulaca* sp.	8 cd
smallflower morningglory	*Jacquemontia tamnifolia* (L.) Griseb.	7 cd
common lambsquarters	*Chenopodium album* L.	6 d
tall morningglory	*Ipomoea purpurea* (L.) Roth	5 de
curly dock	*Rumex crispus* L.	4 def
black medic	*Medicago lupulina* L.	4 def
red morningglory	*Ipomoea coccinea* L.	4 def
annual bluegrass	*Poa annua* L.	2 ef
jimsonweed	*Datura stramonium* L.	2 ef
bermudagrass	*Cynodon dactylon* (L.) Pers.	1 f
dandelion	*Taraxacum officinale* G.H. Weber ex Wiggers	1 f
yellow foxtail	*Setaria pumila* (Poir.) Roemer & J.A. Schultes	1 f
common cocklebur	*Xanthium strumarium* L.	0 f
green foxtail	*Setaria viridis* (L.) Beauv.	0 f
goosegrass	*Eleusine indica* (L.) Gaertn.	0 f
F		10.07
P > *F*		< 0.0001

[z]Values are means of up to 16 replications; means followed by the same letter are not significantly different based on differences of least squares means ($P \leq 0.05$).

Table 4. Number of reniform nematode females developing on seven weed species surveyed in greenhouse test 4.

Common name of weed	Scientific name of weed	Females per g root[z]
common waterhemp	*Amaranthus rudis* Sauer	26 a
broadleaf signalgrass	*Urochloa platyphylla* (Nash) R.D. Webster	11 ab
Pennsylvania smartweed	*Polygonum pensylvanicum* L.	11 ab
giant ragweed	*Ambrosia trifida* L.	7 b
Amazon sprangletop	*Leptochloa panicoides* (J. Presl) A.S. Hitchc.	5 b
bearded sprangletop	*Leptochloa fusca* (L.) Kunth var. *fascicularis* (Lam.) N. Snow	4 b
fall panicum	*Panicum dichotomiflorum* Michx.	0 c
F		7.97
P > *F*		< 0.0001

[z]Values are means of up to 16 replications; means followed by the same letter are not significantly different based on differences of least squares means ($P \leq 0.05$).

yellow nutsedge, junglerice, purple nutsedge, common cocklebur, green foxtail, goosegrass, and fall panicum (Tables 1 through 4).

The average reniform nematode population density in soil associated with the plants examined in the field survey (Tables 5, 6, and 7) exceeded the damage threshold of two nematodes per cm^3 of soil (i.e., 400 nematodes per 200 cm^3 soil) at planting for Mississippi cotton (Wang, 2007). All of the naturally-occurring plants evaluated in the field weed survey supported infection and development of reniform nematode (Table 5). The highest levels of infection occurred on sicklepod, prostrate spurge, hemp sesbania, and velvetleaf. In tests in naturally-infested fields, all of the sown weed species supported infection and development of reniform nematode (Table 6). The highest levels of root infection occurred on sicklepod in the field test. The other four weeds (spurred anoda, velvetleaf, entireleaf morningglory, and teaweed) supported lower levels of root infection than sicklepod but were similar to each other in their response to reniform nematode. Further, levels of root infection on the weeds were often higher than those observed on the cotton plants sown concurrently. Purple and yellow nutsedge plants collected from the field were not good hosts for reniform nematode (Table 7). Nematodes were observed infecting all belowground plant tissues, though roots supported higher numbers of nematodes than either rhizomes or tubers (Table 7). These results are consistent with the levels of infection observed in greenhouse tests (Tables 1 and 2).

Findings from greenhouse experiments to document the host status of weeds identified in the surveys as potentially important hosts for reniform nematode are summarized in Table 8. Reniform nematode population development in inoculated greenhouse tests on nine weed species is compared to susceptible cotton, resistant *G. arboreum* A2-190, and resistant corn in Table 8. When both soil populations and root-associated eggs were considered, velvetleaf, spurred anoda, and hemp sesbania supported as many reniform nematodes as cotton. At the other end of the spectrum, northern jointvetch, common waterhemp, and teaweed supported small reniform nematode populations that were comparable to those that developed on corn and resistant *G. arboreum* A2-190.

DISCUSSION

Methods to reduce reniform nematode populations in cotton excluding extreme chemical treatments remain elusive. Continued efforts to reduce the weeds that support nematodes also should be addressed in order to deprive nematodes of alternative food sources during the cotton production season and in cotton fields rotated to corn or other nonhost crops, or to resistant cultivars of other crops such as soybean. The weed species that support the highest reniform nematode populations should be emphasized in a combined nematode/weed management program. This research identified common groundsel, white clover, sicklepod, hemp sesbania, entireleaf morningglory, velvetleaf, teaweed, and spurred anoda as weeds of particular interest with respect to supporting reniform nematode development and reproduction. Some of these weeds are competitive with cotton and

Table 5. Reniform nematodes associated with seven naturally-occurring weed species in a field survey conducted near Stoneville, MS, in June 2015; the numbers of nematodes collected from soil surrounding the surveyed plants are provided for reference purposes.

Weed species	Females per g root[z]	Reniform nematodes in 200 cm^3 soil
sicklepod	19.5 a	3,464
prostrate spurge	9.2 a	1,286
hemp sesbania	8.6 ab	1,906
velvetleaf	6.6 ab	3,189
purslane	2.9 bc	2,146
Palmer amaranth	1.7 c	1,730
teaweed	1.3 c	687
F	4.25	
$P > F$	0.0009	

[z]Means followed by the same letter are not significantly different at $P \leq 0.05$ based on differences of least squares means.

Table 6. Reniform nematodes associated with roots of five weed species sown in a cotton field naturally-infested with reniform nematode in Stoneville, MS; the mean number of females infecting roots of the susceptible cotton cultivar ST 4946GLB2, and the number of nematodes collected from soil surrounding the test plants are provided for reference purposes.

	Females per g root		Reniform nematodes in 200
Plant	Plant[z]	Cotton	cm^3 soil
sicklepod	961.7 a	31.8	3,907
spurred anoda	61.5 b	24.4	2,020
teaweed	55.2 b	70.9	2,650
entireleaf morningglory	39.7 b	8.5	2,573
velvetleaf	34.7 b	22.5	3,571
F	4.26		
P > *F*	0.0044		

[z]Values are means of 8 replications in two trials; means followed by the same letter are not significantly different based on differences of least squares means ($P \leq 0.05$).

Table 7. Host susceptibility of purple and yellow nutsedge rhizomes, roots and tubers to reniform nematode in a field survey conducted near Stoneville, MS in June 2015; the numbers of nematodes collected from soil surrounding the surveyed plants are provided for reference purposes.

	Females per g of tissue[z]			Reniform nematodes in 200
Weed species	Rhizome	Root	Tuber	cm^3 soil
purple nutsedge	0.05	1.07	0.49 a	1,586
yellow nutsedge	0.05	3.95	0.00 b	1,247

Table 8. Development of reniform nematode populations on nine common Mississippi weeds compared to susceptible cotton cultivar Stoneville 4892 BG/RR, resistant *Gossypium arboreum* accession A_2-190, and resistant corn hybrid Pioneer 3223 in greenhouse tests.

Plant	Nematodes per 100 cm^3 soil[z]	Eggs per 3 g root
cotton	2852.6 a	3555.3 a
velvetleaf	2245.5 a	4303.3 a
entireleaf morningglory	1456.8 ab	340.0 cd
spurred anoda	1110.0 ab	3371.1 ab
hemp sesbania	849.7 abc	5194.2 a
yellow sweetclover	771.0 abc	48.0 e
sicklepod	755.8 abc	769.0 bc
northern jointvetch	387.4 bcd	180.2 cde
common waterhemp	228.2 cd	5.2 f
G. arboreum A2-190	114.2 d	116.9 de
teaweed	112.3 d	167.5 cde
corn	104.5 d	4.5 f
F	5.56	17.84
P > *F*	< 0.0001	< 0.0001

[z]Values are means of 10 observations from two combined trials; means followed by the same letter are not significantly different based on differences of least squares means ($P \leq 0.05$).

may provide a means of sustaining the population of nematodes. Cover crops such as clover, which supported significant reniform nematode levels, may not be the best choice in spite of its capacity to add nitrogen and organic matter to the soil.

Several of the weeds tested in these experiments were evaluated in other locations in the southern United States. The findings of the current study agree with results from tests in Alabama and Georgia (Davis and Webster, 2005; Lawrence *et al*., 2008) that identify sicklepod as an important host for reniform nematode. Further, velvetleaf and entireleaf morningglory were considered important hosts for reniform nematode in two Alabama tests (Dismukes *et al*., 2006; Lawrence *et al*., 2008) and the current work from Mississippi; these weeds were not included in the Georgia study (Davis and Webster, 2005). Consistent results across a broad geographic range suggest that extra efforts to manage these weeds may be needed where reniform nematode populations are present.

The same seed lot was used for sicklepod evaluated in the greenhouse survey and sown adjacent to cotton in a naturally infested field, but the number of females infecting the roots was about 53 times greater in the field test. The reniform nematode density in the soil was higher in the greenhouse test, suggesting that difference in susceptibility within the weed species, differences in aggressiveness in the reniform nematode populations, or perhaps both could be contributing to the observed variability. Additional tests would need to be designed and conducted to determine which of these factors might be responsible for the observed differences.

Nutsedges were not good hosts for reniform nematode in the current trials conducted in Mississippi. All underground parts of the plants appeared to support infection, though it is possible that tuber- and rhizome-associated nematodes were utilizing developing adventitious root tissues that were not yet discernable as true roots to the unaided eye. The poor host status as determined in these experiments agrees with work done in Alabama for purple nutsedge (Dismukes *et al.*, 2006; Lawrence *et al*., 2008) and with work done in Alabama (Lawrence *et al*., 2008) and Georgia (Davis and Webster, 2005) for yellow nutsedge. The consistently poor host status of purple nutsedge in Mississippi tests contradicts a report from Georgia (Davis and Webster, 2005), in which this weed species was identified as an excellent host for reniform nematode in one trial and as a poor host in a second trial. Yellow nutsedge was shown to support reniform and other nematodes in banana fields in Martinique (Quénéhervé *et al*., 2006). This may be an example of infection varying by location and environmental conditions.

Cereal rye and flax did not support reniform nematode infection and development. Similar results were observed in wheat rotation with flax in which *Pratylenchus neglectus* and *P. thornei* populations were reduced and wheat yield was improved (Smiley and Nicol, 2009). However, several other species of nematode were established in nature on flax (Skarbilovich, 1971). The decision to use cereal rye as a cover crop or flax as a rotation crop in fields infested with this pathogen may not increase reniform nematode pressure in the field. In a separate study, Molin and Stetina (2013) found that including a rye cover crop in a cotton production system did not reduce reniform nematode populations in the field.

Several weed species commonly found in production fields in Mississippi have evolved resistance to commonly used herbicides such as glyphosate and acetolactate synthase inhibitors (Heap, 2016). Herbicide resistance increases the likelihood of weed survival and the possibility that these weeds may serve as hosts for reniform nematode. Weeds supporting reniform nematode in this study that may also have been herbicide resistant include water hemp, ragweed, horseweed, and annual bluegrass. As herbicide resistant weeds become more widespread, the contribution of these weeds to maintaining reniform nematode population may be realized.

It should be emphasized that, although the nematode used in the study was from a production cotton field and retained its infectivity on cotton, it is possible that it may exhibit different infectivity towards various weeds. The variability in infection by different reniform nematode biotypes should probably be investigated on a local basis to avoid miscalls as to which plants support or diminish nematode establishment. Local variability among biotypes of both plant and nematode species may be present, which supports or reduces successful establishment in a particular area. Such variability may account for differences in host susceptibility in different regions of the country. Equally important is that weed species are continually being introduced into new regions and their ability to serve as hosts for reniform nematode are largely unknown. For example, Skojac *et al.* (2007) reported 20 new species in the Yazoo-Mississippi Delta region; four of which, *Bowlesia incana*, *Caperonia palustris*, *Eichornia crassipes*, and *Senecio vulgaris*, were weeds of agricultural areas. Field collections in Washington County, MS, a county central to the Yazoo-Mississippi Delta, identified 271 new species in the county and 2 new species to the state (Bryson and Skojac, 2011). The ability of these weeds, such as *Bowlesia incana*, a non-native invasive weed, to

serve as a host for reniform nematode needs to be investigated. *Bowlesia incana* is rapidly spreading across the southern states (Anonymous, 2016) and particularly in no- and reduced-tillage row crop production systems of the Yazoo-Mississippi Delta (Skojac *et al.* 2007). *Bowlesia incana* was observed on the Crop Production Systems research farm in a fallow and soybean field in 2015. The addition of such weeds into an agricultural region necessitates continued diligence in describing the range and diversity of weedy species serving as reniform nematode hosts.

DISCLAIMERS

Mention of trade names or commercial products is solely for the purpose of providing specific information and does not imply recommendation or endorsement by the U. S. Department of Agriculture (USDA). USDA is an equal opportunity provider and employer.

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