

EVALUATION OF COTTON CULTIVARS FOR RESISTANCE AND TOLERANCE TO *ROTYLENCHULUS RENIFORMIS*

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ABSTRACT

Usery, S. R., Jr., K. S. Lawrence, G. W. Lawrence, and C. H. Burmester. 2005. Evaluation of cotton cultivars for resistance and tolerance to *Rotylenchulus reniformis*. *Nematropica* 35:121-133.

Transgenic and conventional cotton cultivars were examined in the greenhouse and two field locations for resistance and tolerance to *R. reniformis*. Greenhouse evaluations of 52 cotton cultivars found all cotton cultivars tested were susceptible to *R. reniformis*. Reproductive factors (Rf) ranged from a high of 59.4 to a low of 4.1. The cultivars SG 105, DP 543 BGII/RR, DP 445BG/RR, and FM 989 R exhibited the lowest 5% of the Rf values. Selected cotton cultivars were evaluated for tolerance to the reniform nematode in both north and south *R. reniformis* infested areas of Alabama. None of the cultivars evaluated were found to be tolerant to *R. reniformis* and able to maintain yield potential in northern AL. In southern AL, ST 4793 R and SG 521 R exhibited greater or equal yields between fumigated and non-fumigated plots; however, neither cultivar was consistently tolerant to *R. reniformis* in northern AL. Results of greenhouse and field testing indicate resistance and consistent tolerance to *R. reniformis* is not currently available. Although there is no consistent tolerance to *R. reniformis*, a high yielding variety suited to the area of production can aid in increased yields in the presence of *R. reniformis*. Results from northern and southern locations in Alabama indicated that cultivars such as DP 451 BR and FM 991 BR, which are intolerant to *R. reniformis*, can be of economic value if nematicides are not implemented.

Key Words: cotton, *Gossypium hirsutum*, resistance, *Rotylenchulus reniformis*, tolerance.

RESUMEN

Usery, S. R., Jr., K. S. Lawrence, G. W. Lawrence, and C. H. Burmester. 2005. Evaluación de la resistencia y tolerancia de cultivares de algodón a *Rotylenchulus reniformis*. *Nematropica* 35:121-133.

Se evaluó la resistencia y tolerancia a *R. reniformis* de cultivares de algodón transgénicos y convencionales en invernadero y en dos localidades en el campo. En las evaluaciones de invernadero de 52 cultivares de algodón se encontró que todos eran susceptibles a *R. reniformis*. Los factores de reproducción (Rf) oscilaron entre 59.4 y 4.1. Los cultivares SG 105, DP 543 BGII/RR, DP 445BG/RR, y FM 989 R exhibieron el 5% más bajo de los valores de Rf. Se evaluó la tolerancia de cultivares seleccionados en áreas infestadas con *R. reniformis* tanto al sur como al norte del estado de Alabama. En el norte de Alabama, no se halló tolerancia al nematodo reniforme o capacidad de mantener la productividad en ninguno de los cultivares evaluados. En el sur de Alabama, los cultivares ST 4793 R y SG 521 R mostraron productividad igual o mayor al comparar lotes fumigados y no fumigados. Sin embargo, ninguno de estos dos cultivares se comportó consistentemente como tolerante a *R. reniformis* en el norte de Alabama. Los resultados de las pruebas de invernadero y de campo indican que ninguno de los cultivares posee resistencia o tolerancia a *R. reniformis*. A pesar de que no se observó una tolerancia consistente a *R. reniformis*, una variedad de alta productividad adaptada a la región puede ayudar a mejorar el desempeño en campos infestados con *R. reniformis*. Los resultados de las localidades en el sur y norte de Alabama, indican que cultivares tales como DP 451 BR y FM 991 BR, que son intolerantes a *R. reniformis*, pueden ser de valor económico si no se aplican nematicidas.

Palabras clave: algodón, *Gossypium hirsutum*, resistencia, *Rotylenchulus reniformis*, tolerancia.

INTRODUCTION

Rotylenchulus reniformis has become the most damaging nematode pest of cotton in the state of Alabama. Since initial identification in 1959, *R. reniformis* has spread rapidly throughout cotton growing regions of the state (Minton and Hopper, 1959; Gazaway and McLean, 2003). It is estimated that *R. reniformis* causes at least a 9% cotton yield loss in Alabama (Blasingame, 2004). Producers currently implement nematicides and crop rotations as their primary management tools against *R. reniformis*. Nematicides such as 1, 3-dichloropropene (1, 3-D) and aldicarb have proven effective at increasing yields but can be prohibitively expensive and offer only short term protection from *R. reniformis* (Lawrence and McLean, 1999). Although crop rotations with non-host crops such as corn and sorghum are effective in reducing populations and damage incurred by *R. reniformis*, rotations with these crops are often economically prohibitive (Davis *et al.*, 2003; Lawrence and McLean, 1999).

The availability of a tolerant and/or resistant cotton cultivar would be a profitable solution to yield reduction caused by *R. reniformis*; however, neither tolerance or resistance has been incorporated into *Gossypium hirsutum* commercial cultivars (Birchfield, 1962; Usery *et al.*, 2005). Resistance and tolerance to plant parasitic nematodes was initially described by Seinhorst in 1966. Resistance is a host response that inhibits the nematode and is measured in terms of the ability of the nematode to reproduce on the host. The relative resistance of a host is assessed by the ratio of the final to the initial population of the nematode (Rf) (Jones *et al.*, 1967; Seinhorst, 1966). If this Rf ratio is less than 1 the nematode density is decreasing and the host is considered resistant. If the ratio is greater than 1 the nematode density is increasing

and the host is susceptible. Thus some intermediate level of reproduction above the Rf value of 1 but at a level below a standard susceptible cultivar can be considered moderately or partially resistant (Boerma and Hussey, 1992). Tolerance is the ability of the host to survive and produce satisfactory yields when parasitized by a nematode at a level that causes economic yield losses to other cultivars. Like resistance, tolerance is a relative concept often comparing yields between nematicide treated and non-treated cultivars. Tolerance and resistance are considered independent characteristics (Trudgill, 1991) and not necessarily related, although both can be beneficial.

In 1988, Jones *et al.*, registered four cotton germplasm lines as resistant to *R. reniformis*. The Rf reproduction ratio was not less than one; however, these lines produced only 21 to 36% of the eggs/g of root compared to the standard Deltapine 41 cultivar. Cook *et al.*, (1997) accepted three breeding lines as resistant to *R. reniformis* when the Rf was statistically less than the standard susceptible cultivar, Deltapine 16, even though the Rf values on all tested lines were greater than 50. Robinson and Percival (1997) examined 46 accession of *G. hirsutum* for resistance and determined none supported less reproduction of *R. reniformis* than the standard cultivar Deltapine 16. Further research examining resistance and tolerance of five Texas cotton breeding lines in North Carolina found all which supported lower levels of *R. reniformis* than the standard cultivars Deltapine 50 and Stoneville LA 887, although the breeding lines had higher tolerance indices (Koenning *et al.*, 2000). Tolerance to plant-parasitic nematodes is largely influenced by location and environmental conditions (Soriano *et al.*, 2000). Multiple studies have determined variations in cotton yield as affected by *R. reniformis* varied by location and over years (Koenning *et al.*,

2000; Davis and May, 2003; Usery *et al.*, 2005). Regression analysis of cotton yield as a dependent variable of *R. reniformis* numbers often have low r^2 values indicating nematode numbers account for only a small part of the observed variability in treatments (Wallace, 1987).

The current study focuses on identification of possible resistance and/or tolerance among currently available transgenic and non-transgenic cotton cultivars in Alabama. Selection of cultivars for testing was based on availability to Alabama cotton producers and suitability to the growing region.

MATERIALS AND METHODS

Greenhouse Trials

Greenhouse evaluations were conducted at the Plant Science Research Center of the Alabama Agricultural Experiment Station on the campus of Auburn University, Auburn, AL. Fifty-two transgenic and non-transgenic cotton cultivars and lines were evaluated for host suitability to *R. reniformis* and early season growth parameters.

Inoculum Preparation

The culture of *R. reniformis* was procured by collecting samples of soil from various infested field locations throughout Alabama. *Rotylenchulus reniformis* was propagated and increased on cotton (PM 1218 BR) in the greenhouse. After sixty days, cotton plants were removed and the nematodes were extracted from the soil by combined gravity screening and sucrose centrifugal flotation (Jenkins, 1964). *Rotylenchulus reniformis* were standardized to 1000 juveniles and vermiform adults per 3 ml of water for inoculation.

Host Suitability

Fifty-two cultivars were grown in 150 cm³ Conetainer® filled with a loamy sand

soil (72.5% sand, 25% silt, 2.5% clay, pH 6.4). The soil was autoclaved twice at 121°C and 103.4 kPa for two hours on two consecutive days. Seeds were planted and allowed to germinate and grow for seven days, at which time a standardized suspension of 1,000 *R. reniformis* juveniles and vermiform adults were pipetted into each container.

Greenhouse experiments were arranged in a randomized complete block design with five replications and the test was conducted three times. Sixty days after inoculation, plants were harvested, nematodes were extracted from the soil, and eggs were removed from the roots. *Rotylenchulus reniformis* was extracted from the soil as previously described. Eggs were extracted from the roots by shaking for 4 min. in 0.6% sodium hypochlorite (NaOCl) solution (Hussey and Barker, 1973). After enumeration of *R. reniformis* vermiform stages and egg populations, reproductive factor values (Rf = final population/initial population) were determined.

Early Season Growth Parameters

Fifty-two cotton cultivars were planted and inoculated using the methods previously described. Twenty-days following inoculation, plants were harvested. Plant height and fresh weight were obtained and cotton shoots were dried at 80.0°C in an oven for 48 hr and weights were recorded. Roots were washed to remove all soil and foreign matter. Root architecture was assessed by determining total root length, root diameter, root surface area, root projected area, root volume, and number of root tips. These analyses were obtained by utilizing WinRhizo software version 5.0 (Regent Instruments, Inc., Quebec, Canada) with a Hewlett Packard scanner (HP ScanJet 6100C). Root fresh weights and dry weights were also collected.

Field Evaluation of Tolerance

Field experiments were conducted throughout 2002-2004 in north and south Alabama with cultivars varying in each location and over time. The north Alabama test was located in a producer's field in Limestone County that was naturally infested with *R. reniformis*. The soil was classified as silt loam (23.7% sand, 53.7% silt, 22.5% clay, pH 6.5). Plots were conventionally tilled in all years. Cultivars were compared with and without a nematicide in all years. In 2002, 5.7 kg/ha (0.85 kg a.i./ha) of aldicarb were applied to nematicide treated plots. Aldicarb was applied at planting in the seed furrow with chemical granular applicators attached to the planter. In 2003 and 2004, 12.9 L/ha of 1, 3-D were applied one month before planting. The 1, 3-D was applied using a modified John Deere ripper/bedder injection device. A CO₂ charged system was used to propel the fumigants through flow regulators mounted on stainless steel delivery tubes attached to the trailing edge of forward-swept chisels. The 1, 3-D was injected 39 cm. deep, and immediately bedded with disk tillers to seal the chisel trace. Plots not treated with 1, 3-D were ripped, bedded, and conventionally tilled. Disulfoton was used to control insects in non-aldicarb plots in 2002 and in all plots in 2003 and 2004. Plots consisted of 2 rows, 7.62 m long with 101 cm row spacing in all years. The 2002 test was arranged as paired plots in a randomized complete block design with four replications. The 2003 and 2004 tests were arranged in a split-plot in a randomized complete block design with four replications. In all years, plots were sampled for *R. reniformis* populations at planting, mid-season, and maturity. Twenty 2.5-cm-diam. soil cores were collected to 20 cm-depth from both rows in a plot using a systematic sampling pattern. Composite soil samples

were sealed in plastic bags and stored in a cooled ice chest as they were collected, then transferred to a 5°C refrigerator for storage prior to extraction. *Rotylenchulus reniformis* was extracted from a 150-cm³ sub-sample and enumerated using the methods previously described. In 2004, all plots were evaluated for seedling survival, plant height and early season vigor at the two true leaf growth stage. Plant heights were measured from a 5 plant sub-sample per plot and measurements were taken from the cotyledonary nodes to the apical tip. A visual vigor rating on a 1-5 scale, with 1 being the least vigorous and 5 being the most vigorous, was performed. Plots were harvested at maturity and seed cotton weights were recorded for each plot in all years.

Thirty-two cotton cultivars were examined with and without the nematicide 1, 3-D in south Alabama in Escambia County. The southern Alabama field was a sandy loam (65% sand, 25% silt, 9.75% clay, pH 6.0), naturally infested with *R. reniformis*. The 1, 3-D was injected at 12.9 L/ha using the methods previously described one month prior to planting. Plots not treated with 1, 3-D were ripped, bedded, and conventionally tilled. Disulfoton was used to control insects in all plots. Plots consisted of one row, 7.62 m long with a 96.5 cm. row spacing and were arranged as a split-plot in a randomized complete block design with six replications. All plots were sampled for *R. reniformis* populations at planting and maturity. Samples were extracted and *R. reniformis* populations were enumerated using the methods previously described. Seed cotton was weighed and recorded at harvest.

All data were subjected to analysis of variance using general linear model procedure (GLM) of the Statistical Analysis System (SAS Institute, Cary, NC), and means were separated with Fisher's protected least significant difference test ($P \leq 0.05$).

Meaningful paired comparison t-tests were used to determine significant differences between nematicide treated and untreated varieties. Pearson's correlation coefficients were utilized to determine linear associations between parameters measured. All levels of significance reported herein are at the $P \leq 0.05$ level unless otherwise stated.

RESULTS

Host Suitability

All cotton cultivars tested supported *R. reniformis* reproduction. Data from the repeated tests were combined for analysis because a significant interaction between tests was not observed. Total vermiform *R. reniformis* extracted from the soil ranged from a high of 20,487/150 cm³ of soil on FM 958 LL to a low of 520 on DP 543 BGII/RR (Table 1). Egg production/root system varied from 51,325 to 2,446 on BCG 50 R and SG 105, respectively. Lower numbers of eggs/g of dry root were produced on SG 105 and DP 543 BGII/RR as compared to all other cultivars. The Rf values for all cotton tested ranged from 59.4 to 4.1. The relative frequency of nematode reproduction was symmetrical, producing a normal distribution with 90% of the cultivars producing total population numbers between 11,000 and 49,000. Only 10% of the cultivars tested produced total populations below 10,000 or above 50,000. SG 105, DP 543 BGII/RR, DP 445 BG/RR, and FM 989 R produced lower Rf values and had fewer eggs/g of root than 95% of the cultivars tested.

Early Season Growth Parameters

Root architecture analyses measuring total root length, root surface area, projected area, root volume, number of root tips, and average root diameter were deter-

mined for selected cultivars (Table 2). ST 5599 BR produced the largest root system as measured for each parameter. ST 5599 BR formed a greater root volume and total root projected area as compared to 82% and 76% of cultivars tested, respectively. Seventy percent of cultivars tested had a lower surface area and total number of root tips while 52% produced shorter roots with smaller diameters than ST 5599 BR. Significant Pearson's correlation coefficients indicated a negative relationship between root length, surface area, and total projected area of the root architecture and population levels of *R. reniformis* vermiform stages at 60 DAP. Root parameters were not influenced by the number of eggs produced.

Field Evaluation of Tolerance

In the 2002, in the Limestone Co. field trial there were no differences in seed cotton yields between plots treated and not treated with aldicarb (data not shown). The application of aldicarb produced no effect on *R. reniformis* populations.

In 2003 and 2004, cultivars differed in their responses to the application of 1,3-D by location and over time as measured by seed cotton yield and *R. reniformis* numbers. In 2003 in Limestone Co., seed cotton yields for DP 451BR and ST 4793R were not increased with the nematicide, indicating potential tolerance (Table 3). DP 451 BR produced more seed cotton than 41% of the cultivars in untreated plots. The sum of initial, mid season, and harvest vermiform stage *R. reniformis* numbers ranged from 2,616/150 cm³ for DP 444 BR to 614 for DPL 451 BR in the 1,3-D treated plots. The application of 1,3-D increased seed cotton yields overall by 14% and reduced *R. reniformis* numbers by 8%. Pearson's correlations coefficients indicated seed cotton production was influenced by variety ($P = 0.001$; $R^2 = -0.404$)

Table 1. Greenhouse evaluations of 52 cotton cultivars for *Rotylenchulus reniformis* reproduction measured by numbers of vermiform stages and eggs per root system, and the reproduction factor, and plant growth parameters measured by shoot dry weight and root dry weight.

Variety	<i>Rotylenchulus reniformis</i> ^s	<i>Rotylenchulus reniformis</i> eggs	Rf value ^t
SG 105	1,646 i-k	2,446 p	4.1
DP 543 BGII/RR	520 k	4,252 op	4.8
DP 445 BG/RR	595 k	5,264 n-p	5.9
FM 989 R	833 jk	6,682 m-p	7.5
DP 5690 R	2,124 h-k	9,154 l-p	11.3
ST 4793 R	3,361 f-k	8,285 l-p	11.6
STX 6848 R	1,151 jk	11,225 j-p	12.4
SG 747	2,441 g-k	10,480 k-p	12.9
SG 215 BR	3,546 f-k	10,477 k-p	14.0
ST 5599 BR	2,078 h-k	14,160 i-p	16.2
STX 3636 B2R	1,715 i-k	16,029 h-p	17.7
STX 4575 BR	2,472 g-k	17,266 g-p	19.7
SG 521 R	1,840 i-k	17,974 g-p	19.8
DP 432 R	2,673 g-k	17,582 g-p	20.3
DP Deltapearl	4,929 d-k	18,849 f-o	23.8
FM 991 R	5,122 c-k	18,669 f-o	23.8
PM 1218 BR	7,802 c-g	16,429 h-p	24.2
BCG 28 R	4,738 e-k	2,0291 e-n	25.0
DP 434 R	4,574 e-k	20,832 e-n	25.4
DP 555 BR	2,001 h-k	23,828 e-l	25.8
ST 5303 R	5,622 c-k	21,012 e-m	26.6
DP 494 R	5,817 c-k	22,017 e-m	27.8
BCG 24 R	1,978 h-k	25,879 d-k	27.9
STX 4686 R	2,943 g-k	26,072 d-j	29.0
STX 5454 B2R	2,804 g-k	26,729 d-j	29.5
STX 6636 BR	4,581 e-k	25,161 d-k	29.7
ST 4892 BR	10,066 b-e	19,856 e-n	29.9
FM 966 LL	3,811 f-k	26,858 d-i	30.7
FM 960 BR	3,438 f-k	27,424 d-i	30.9
DP 493	4,612 e-k	27,115 d-i	31.7
DP 451 BR	3,925 f-k	28,093 d-i	32.0
DP 458 BR	6,034 c-k	28,712 d-i	34.7
ST 5242 BR	4,319 f-k	30,952 d-h	35.3
FM 989 BR	4,604 e-k	30,720 d-h	35.3

^sPopulations per 150 cm³ of soil.

^tRf = final vermiform nematodes and eggs/initial population. Means within columns followed by different letters are different according to Fisher's protected least significant difference test ($P \leq 0.05$).

Table 1. (Continued) Greenhouse evaluations of 52 cotton cultivars for *Rotylenchulus reniformis* reproduction measured by numbers of vermiform stages and eggs per root system, and the reproduction factor, and plant growth parameters measured by shoot dry weight and root dry weight.

Variety	<i>Rotylenchulus reniformis</i> ^s	<i>Rotylenchulus reniformis</i> eggs	Rf value ^t
DP 449 BR	15,280 ab	20,111 e-n	35.4
FM 991 B2R	6,798 c-i	29,123 d-i	35.9
FM 960 R	2,094 h-k	34,067 b-f	36.2
DP 444 BR	10,344 b-d	26,085 d-j	36.4
DP 424 B2R	8,490 c-f	28,300 d-i	36.8
DP 5415 R	7,409 c-h	30,214 d-h	37.6
FM 991 BR	10,626 bc	27,066 d-i	37.7
DP 393	3,623 f-k	34,067 b-f	37.7
DPLX 02T57R	4,427 f-k	34,029 b-f	38.5
DP 436 RR	6,273 c-j	32,316 c-g	38.6
PHY 410 R	5,670 c-k	34,631 b-e	40.3
FM 958 LL	20,487 a	26,291 d-j	46.8
DP 491	18,834 a	28,145 d-i	47.0
FM 960 B2R	2,457 g-k	46,543 a-c	49.0
DP 455 BG/RR	4,015 f-k	48,050 ab	52.1
ST 4646 B2R	5,524 c-j	47,535 a-c	53.1
BCG 50 R	3,909 f-k	51,325 a	55.2
DP 488 BR	19,699 a	39,655 a-d	59.4
LSD ($P \leq 0.05$)	5,528	15,582	—

^sPopulations per 150 cm³ of soil.

^tRf = final vermiform nematodes and eggs/initial population. Means within columns followed by different letters are different according to Fisher's protected least significant difference test ($P \leq 0.05$).

and nematicide, ($P = 0.002$; $R^2 = -0.307$). Yield was also influenced by the initial ($P = 0.063$; $R^2 = -0.191$), midseason ($P = 0.035$; $R^2 = -0.216$), and total nematode ($P = 0.071$; $R^2 = -0.185$) populations.

In 2004, seed cotton yields were increased from 2% to 19% over all cultivars tested in plots treated with 1,3-D. Seed cotton yields varied from a high of 5,951 kg/ha for ST 5599 BR treated with 1, 3-D to a low of 4,067 for ST 4793 R untreated (Table 4). The lowest response to 1,3-D was by FM 989 R with a seed cotton increase of 2.1%. Although possible tolerance was observed for ST 4793 R and DP 451 BR in 2003, seed

cotton yields were increased with 1,3-D for ST 4793 R and DP 451 BR in 2004. Season total vermiform stage numbers ranged from 3,721 to 817/150 cm³ for ST 4892 BR and FM 682 R, respectively. Pearson's correlation coefficients indicated *R. reniformis* population levels throughout the season affected 54% of the variation in seed cotton yields observed ($P < 0.0001$; $R^2 = -0.539$). Seed cotton yields were also correlated with plant vigor ratings taken at the second leaf stage. Thirty-two percent of the variation in yield was associated with vigorous seedling plant growth ($P = 0.029$; $R^2 = 0.323$). Plants of DP 444 BR, DP 451 BR, PM 1218BR, and ST

Table 2. Greenhouse evaluations of 17 cotton cultivars root architecture as measured by total root length, root surface area, projected area, root volume, number of root tips and average root diameter.

Variety	Total root length cm	Total root surface area cm ²	Total projected area cm ²	Total root volume cm ³	Average root diameter mm	Total number of root tips
ST 5599 BR	326.5 a	153.3 a	49.128 a	5.97 a	1.498 a	333.6 a
FM 989 BR	312.7 ab	126.0 a-d	40.118 a-d	4.09 b-d	1.284 a-d	257.2 a-d
DP 5415 R	305.0 a-c	128.2 a-c	40.816 a-c	4.43 a-c	1.339 a-c	302.2 ab
FM 960 BR	304.2 a-c	102.9 b-f	32.756 b-g	2.80 c-f	1.067 de	269.6 a-c
DP 444 BR	302.9 a-c	135.7 ab	43.202 ab	4.98 ab	1.430 ab	222.2 b-d
DP 451 BR	294.5 a-d	130.8 a-c	36.512 b-e	3.61 b-e	1.236 b-e	219.8 b-d
PM 1218 BR	275.8 a-e	87.8 d-f	31.758 d-h	2.90 c-f	1.148 c-e	211.8 cd
FM 989 R	266.4 a-f	111.0 c-e	35.344 b-f	3.80 b-d	1.340 a-c	227.8 b-d
STX 4686 R	250.0 b-f	87.7 d-f	27.924 e-h	2.48 d-f	1.105 c-e	264.6 a-c
DP 458 BR	248.7 c-f	93.5 c-f	29.750 d-h	2.89 c-f	1.159 c-e	217.2 cd
SG 215 BR	234.9 d-f	93.9 c-f	29.880 d-h	3.02 c-f	1.267 a-d	230.0 b-d
FM 991 BR	232.2 d-f	86.9 d-f	27.676 e-h	2.63 d-f	1.194 b-e	241.0 b-d
ST 4892 BR	226.3 e-f	102.4 b-f	29.414 e-h	3.14 c-f	1.305 a-d	178.2 d
DP 555 BR	224.6 e-f	95.0 c-f	30.220 c-h	3.20 c-f	1.347 a-c	194.0 cd
DP 449 BR	222.1 e-f	77.1 ef	24.552 gh	2.07 ef	1.070 de	222.6 b-d
ST 4793 R	217.5 e-f	68.6 f	21.838 h	1.73 f	0.996 e	212.2 cd
DP 436 R	210.5 f	80.2 ef	25.532 fh	2.45 d-f	1.175 c-e	207.0 cd
LSD (P ≤ 0.05)	63.2	39.1	10.657	1.6512	0.2482	83.343

Means within columns followed by different letters are different according to Fisher's protected least significant difference test ($P \leq 0.05$).

5599 BR displayed the highest vigor ratings while the least vigorous cultivar was DP 5415 R. The total *R. reniformis* population levels were negatively correlated with seedling plant vigor ($P = 0.016$; $R^2 = -0.244$).

Thirty-two cultivars were evaluated in Escambia Co. in southern Alabama. The cotton cultivars differed in their responses to 1,3-D as measured by seed cotton yield and *R. reniformis* populations. Seed cotton yields in the nematicide treated plots were increased ($P \leq 0.10$) for 72% of the cultivars tested (Table 5). Seed cotton yields ranged from 3,238 to 966 kg/ha for FM 991 BR and DP 436 R, respectively. Cultivars ST 4793 R and SG 521 R were the only ones that produced higher yields in the non-

fumigated plots as compared to the fumigated plots, indicating potential tolerance, although FM 991 BR produced 428 and 816 kg/ha more seed cotton in non-fumigated plots than ST 4793 R and SG 521 R, respectively. Pearson's coefficients determined a significant relationship between seed cotton yield and nematode populations ($P = 0.005$; $R^2 = -0.143$), although only 14% of the variation in yield was influenced by the nematode population.

DISCUSSION

Results from the greenhouse and the field reveal that resistance to *R. reniformis* in transgenic and non-transgenic cotton

Table 3. *Rotylenchulus reniformis* end of season total populations and seed cotton yields in cotton cultivar evaluations conducted in northern Alabama in 2003.

Variety	Season total <i>R. reniformis</i> populations* 1,3-D [†] treated	Season total <i>R. reniformis</i> populations untreated	Seed cotton kg/ha 1,3-D treated [‡]	Seed cotton kg/ha untreated	% Yield increase from 1,3-D [†]
DP 5415 R	1,470 bc	1,363 c	4,900 b	4,777 bc	2.6
ST 4892 BR	2,018 b	739 d	4,718 bc	4,480 c	5.3
DP 451 BR	614 d	1,292 cd	5,433 a	5,497 a	-1.2
DP 436 R	1,077 cd	1,782 bc	5,176 ab	4,956 b	4.4
DP 444 BR	2,616 ab	1,569 bc	5,835 a	5,150 ab	13.3*
DP 449 BR	1,014 c	1,408 bc	5,310 a	4,986 b	6.5
PM 1218 BR	1,393 cd	1,383 c	5,236 ab	4,446 c	17.8*
SG 215 BR	1,084 d	1,234 cd	5,314 a	4,889 b	8.7
PM 1199 R	833 d	1,204 cd	4,404 c	4,118 c	6.9
ST 4793 R	627 d	700 d	5,035 b	5,050 b	-0.3
ST 5599 BR	1,065 d	3,205 c	5,977 a	5,150 ab	16.1**
FM 989 BR	1,123 cd	1,512 c	5,518 a	4,848 b	13.8
LSD (P ≤ 0.05) [‡]		921		679	

Means within columns followed by different letters are different according to Fisher's protected least significant difference test (P ≤ 0.05).

*Populations per 150 cm³ of soil.

[†]1,3-D = 1,3-dichloropropene.

[‡]Single and double asterisks indicate percent yield increase due to 1,3-dichloropropene as compared to untreated at (P ≤ 0.10) and (P ≤ 0.05), respectively.

[§]Significant interactions between cultivars and nematicide indicated the factors are not independent and thus are compared with a single LSD value.

cultivars and lines used in Alabama is currently unavailable. Our greenhouse results agree with past findings that the cotton cultivars of *G. hirsutum* currently available are comparable with older cultivars and are susceptible to *R. reniformis*, allowing the nematode to increase population densities (Birchfield, 1962; Usery *et al.*, 2005).

While statistical differences existed between total population development of *R. reniformis* life stages and eggs, no cultivar tested exhibited an Rf value lower than 1.0. The 52 cotton cultivars and lines tested were all susceptible to *R. reniformis*. Previous researchers have identified resistance

as a percentage of reproduction compared with a standard susceptible control cultivar that is arbitrarily chosen. Often in these cases, reproduction has been reported to be reduced 50% to 90%, imparting a statistical difference between the arbitrarily selected standard susceptible control and the cultivars or lines evaluated, thus a resistant rating is applied (Cook *et al.*, 1997; Davis and May, 2003). However, when Rf values are determined, the reproduction potential is well over the Rf minimum value of 1.0, indicating the evaluated lines or cultivars are actually susceptible and not resistant to the nematode. The resistant

Table 4. *Rotylenchulus reniformis* end of season total populations and seed cotton yields in cotton cultivar evaluations conducted in northern Alabama in 2004.

Variety	Season total <i>R. reniformis</i> populations* 1,3-D [†] treated	Season total <i>R. reniformis</i> populations untreated	Seed cotton kg/ha 1,3-D treated [‡]	Seed cotton kg/ha untreated	% Yield increase from 1,3-D [‡]
DP 444 BR	1,944 a-c	3,605 a	5,460 ab	4,808 a-c	13.6
DP 449 BR	2,085 a-c	3,025 a-c	4,679 c-d	4,319 cd	8.3
DP 451 BR	3,264 ab	3,347 ab	5,425 a-c	5,047 ab	7.5
DP 5415 R	1,564 a-c	1,873 a-c	4,385 d	4,233 cd	3.6
FM 960 BR	1,467 a-c	3,373 ab	5,421 a-c	5,161 a	5.0
FM 989 R	682 c	3,257 ab	5,084 b-d	4,979 ab	2.1
FM 991 BR	1,011 bc	3,263 ab	4,838 b-d	4,734 a-c	2.2
PM 1218 BR	2,336 a-c	2,626 a-c	4,959 b-d	4,534 b-d	9.4
ST 4793 R	1,866 a-c	3,296 ab	4,857 b-d	4,067 d	19.4*
ST 4892 BR	2,008 a-c	3,721 a	5,012 b-d	4,599 a-d	9.0
ST 5599 BR	1,255 a-c	2,221 a-c	5,951 a	5,057 ab	17.7**
STX 4686 R	1,615 a-c	2,485 a-c	5,255 a-c	5,028 ab	4.5
LSD (P ≤ 0.05) [‡]		2,554		785	

Means within columns followed by different letters are different according to Fisher's protected least significant difference test (P ≤ 0.05).

*Populations per 150 cm³ of soil.

[†]1,3-D = 1,3 dichloropropene.

[‡]Single and double asterisks indicate percent yield increase due to 1,3 dichloropropene as compared to untreated at (P ≤ 0.10) and (P ≤ 0.05), respectively.

[§]Significant interactions between cultivars and nematicide indicated the factors are not independent and thus are compared with a single LSD value.

rating applied is completely dependent on the reproductive potential of the arbitrarily selected susceptible control. The reproduction levels, although less than the susceptible cultivar, would likely sustain nematode population densities at economically damaging levels to cotton.

We theorized that larger vigorously growing seedling cotton cultivars would also have larger root systems that could better support the host plant in the presence of *R. reniformis* populations. *Rotylenchulus reniformis* population densities in this study were negatively correlated with seedling plant vigor. Fast growing vigorous cotton cultivars were able to tolerate

R. reniformis populations better than the slower growing cultivars. Field plots confirmed that yield was also positively correlated with vigorous cotton seedling growth. Interestingly, ST 5599 BR produced the largest root system in the WinRhizo evaluation and was neither resistant nor tolerant to *R. reniformis* in our tests.

Due to the inconsistent performance across locations over three years of this study, we confirmed with previous studies that tolerance to *R. reniformis* has not been incorporated into the currently available cotton cultivars (Cook *et al.*, 1997; Robinson and Percival, 1997; Robinson *et al.*, 1997). In Limestone Co, the *R. reniformis*

Table 5. Evaluations of cotton cultivar tolerance to *Rotylenchulus reniformis* as measured by *R. reniformis* populations and seed cotton yields conducted in south Alabama, in 2003.

Variety	Harvest <i>R. reniformis</i> populations ^w 1,3-D ^y treated	Harvest <i>R. reniformis</i> populations untreated	Seed cotton kg/ha 1,3-D ^x treated	Seed cotton kg/ha untreated	% Yield increase from 1,3-D treated ^y
BCG 28 R	1,158 bc	1,236 bc	1,831 j-l	1,400 h-l	30.8
DP Delta Pearl	1,236 bc	850 bc	2,704 b-e	2,058 c-g	31.4**
DP 436 R	695 bc	875 bc	2,161 g-k	966 l	123.7**
DP 444 BR	953 bc	1,854 abc	1,790 kl	1,574 e-k	13.7
DP 448 B	1,390 bc	4,171 a	2,681 c-f	1,416 h-l	89.3**
DP 449 BR	2,163 abc	1,545 bc	2,423 d-i	1,929 d-h	25.6*
DP 451BR	927 bc	1,468 bc	2,174 g-k	1,345 i-l	61.6*
DP 458 BR	1,236 bc	1,313 bc	2,730 b-e	1,540 f-k	77.3**
DP 491	334 c	1,545 bc	2,363 d-i	1,059 j-l	123.1**
DP 493	1,004 bc	2,008 abc	2,241 f-k	1,841 d-i	21.7
DP 5415 R	1,622 bc	1,931 abc	2,508 d-g	1,574 e-k	59.3*
DP 555 BR	1,957 abc	2,086 abc	3,156 ab	2,629 ab	20.0*
DP 5690 R	1,931 abc	2,085 abc	2,700 b-f	2,120 b-e	27.4**
DP 33 B	772 bc	3,090 ab	2,753 b-d	1,811 d-i	52.0**
FM 991R	1,390 bc	1,545 bc	3,018 a-c	2,868 a	5.2
FM 958 B	1,159 bc	2,472 abc	2,319 d-i	1,485 h-l	56.2**
FM 960 BR	1,390 bc	2,703 abc	2,274 e-j	1,778 e-i	27.9*
FM 966	1,081 bc	2,240 abc	1,970 i-l	1,459 h-l	35.0*
FM 989	1,236 bc	1,622 bc	2,199 g-k	1,341 i-l	64.0**
FM 989 BR	1,081 bc	2,240 abc	2,379 d-i	1,287 i-l	84.8**
FM 991BR	1,159 bc	1,004 bc	3,238 a	2,971 a	9.0
PM 1218 BR	1,158 bc	1,390 bc	2,192 g-k	1,573 e-k	39.4**
PHY 410 R	618 c	1,236 bc	2,184 g-k	1,012 kl	115.8**
PHY 510 R	1,159 bc	1,390 bc	2,530 d-g	1,702 d-i	48.6**
ST 4793 R	2,394 abc	1,313 bc	2,149 g-k	2,543 a-c	-15.5
ST 4892 BR	1,236 bc	1,236 bc	2,034 h-l	1,518 g-l	34.0*
ST 5303 R	541 c	1,777 abc	2,469 d-h	1,381 h-l	78.8**
ST 5599 BR	1,236 bc	1,468 bc	2,045 h-l	1,457 h-l	40.4**
STX 0203 BR	1,339 bc	1,004 bc	1,685 l	1,370 h-l	23.0
SG 215 BR	566 c	2,008 abc	2,387 d-i	2,096 b-f	13.9
SG 521 R	1,931 abc	1,236 bc	2,116 g-l	2,155 b-d	-1.8
SG 747	1,004 bc	2,086 abc	2,391 d-i	1,609 d-j	48.6**

LSD ($P \leq 0.05$)^z

2,467

563.16

Means within columns followed by different letters are different according to Fisher's protected least significant difference test ($P \leq 0.05$).

^wPopulations per 150 cm³ of soil.

^y1,3-D = 1,3 dichloropropene.

^zSingle and double asterisks indicate percent yield increase due to 1,3 dichloropropene as compared to untreated at ($P \leq 0.10$) and ($P \leq 0.05$), respectively.

^zSignificant interactions between cultivars and nematicide indicated the factors are not independent and thus are compared with a single LSD value.

populations in 2002 were extremely high; however, seed cotton yield differences between cultivars treated with a nematicide and untreated cultivars were not observed. The definition of tolerance (Seinhorst, 1966) would indicate all cultivars tested were tolerant since yields were not different between nematicide treated and non-treated plots; however, we believe this could not feasibly be the case. Previous studies in this county have shown aldicarb is susceptible to enhanced microbial degradation (McLean and Lawrence, 2003; Lawrence *et al.*, 2005). Soil from our location was analyzed for enhanced aldicarb microbial degradation and it was determined that aldicarb was completely degraded within 10 days, thus was not effective in reducing *R. reniformis*. Due to the microbial degradation of aldicarb, differences between nematicide treated and untreated cultivars were undetectable in 2002.

Rotylenchulus reniformis populations in 2002 were lower than in 2003; however, seed cotton yield differences ($P \leq 0.05$) existed between 1,3-D treated cultivars and untreated cultivars. Tolerance was tentatively identified in DPL 451BR and ST 4793 R because the yield of these cultivars was not increased by the addition of the nematicide. In 2004, no variety performed as well as its nematicide treated counterpart. The higher populations of *R. reniformis* and reduced rainfall in 2004 probably increased the disease pressure of the nematode and stress placed on the plants. Pathogenicity and reproductive potential of *R. reniformis* is affected by the environmental conditions (Bird, 1983) which would also affect tolerance. Consistent tolerance to *R. reniformis* was not observed across three years of testing. Although tolerance was not constant over time, the increased yield of DP 451 BR in the absence of a nematicide indicates it should

be an economically profitable cultivar in the northern Alabama growing region.

Although tests were conducted for three years in Escambia Co., hurricanes in 2002 and 2004 prevented collection of seed cotton yields in those years. In 2003, *R. reniformis* pressure in the southern location was sufficient to decrease ($P \leq 0.05$) seed cotton yields in the plots not treated with 1,3-D. Cultivars ST 4793 R and SG 521 R exhibited greater or equal yields when not treated with the nematicide which could indicate a degree of tolerance for this area. While the positive performance by ST 4793 R and SG 521 R seems to indicate tentative tolerance to *R. reniformis*, the inability to test these cultivars over multiple years at the same location, and poor performance in northern Alabama, does not allow labeling these two cultivars tolerant. While ST 4793 R and SG 521 R indicated provisional tolerance, FM 991 BR produced the highest yield in the untreated plots, which indicates FM 991 BR could be the most profitable cultivar for the producers in south Alabama at this time.

In conclusion, our results of greenhouse and field testing at two Alabama locations indicate true resistance and tolerance to *R. reniformis* is not present in the cultivars evaluated. In addition, although there is no available tolerance to *R. reniformis*, use of a high yielding variety suited to the area of production can aid in increased yields in the presence of *R. reniformis*. Results from northern and southern locations in Alabama indicated that cultivars such as DP 451 BR and FM 991 BR, which are intolerant to *R. reniformis*, can be of economic value if nematicides are not implemented.

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