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Effects of *Meloidogyne arenaria* infection on *M. incognita*-resistance in tobacco

Thomas J. BAUM*+, Bruce A. FORTNUM** and Stephen A. LEWIS*

* *Department of Plant Pathology and Physiology, Clemson University, Clemson, SC 29634-0377, USA, and*
** *Pee Dee REC, Clemson University, Florence, SC 29501-9603, USA.*

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Summary – Roots of tobacco resistant to *Meloidogyne incognita* host races 1 and 3 were separated into two portions using three different split-root techniques. One portion of each plant root was inoculated with eggs of *M. arenaria* and the other root portion with *M. incognita*. Eggs of *M. arenaria* and *M. incognita* were applied to separate root portions either simultaneously or *M. incognita* inoculation was delayed. After 45 to 60 days in the greenhouse, root portions were rated for galling and egg mass number. *M. arenaria* infection of one root portion did not systemically predispopse the other portion to *M. incognita* infection. Plants with intact root systems were maintained at 25, 28, 31, or 35 °C and inoculated simultaneously with eggs of both species. *M. incognita*-resistance failed at temperatures above 28 °C. *M. arenaria* infection did not alter the *M. incognita*-resistance phenotype at different temperatures.

Résumé – ***Influence de l'infestation par* Meloidogyne arenaria *sur la résistance à* M. incognita *chez le tabac*** – Le système racinaire de plants de tabac résistants à *Meloidogyne incognita* races 1 et 3 est séparé en deux parties en utilisant trois techniques différentes. Une partie est inoculée avec des œufs de *M. arenaria* et l'autre avec des œufs de *M. incognita*. Les œufs de *M. arenaria* ou de *M. incognita* sont placés sur l'une ou l'autre partie du système racinaire, soit simultanément, soit successivement, mais en décalant l'inoculation de *M. incognita*. Après 45 à 60 jours de croissance en serre des plants infestés, l'importance de l'infestation et le nombre de masses d'œufs sur les deux parties du système racinaire sont évalués. L'infestation d'une partie des racines par *M. arenaria* ne prédispose pas systématiquement l'autre partie à l'infestation par *M. incognita*. Des plants de tabac résistants à *M. incognita*, à système racinaire intact, sont inoculés simultanément avec des œufs des deux espèces et maintenus à 25, 28, 31 ou 35 °C. La résistance à *M. incognita* disparaît aux températures supérieures à 28 °C. Quelle que soit la température, l'infestation par *M. arenaria* n'a pas d'effet sur le phénotype résistant à *M. incognita*.

Key-words : *Meloidogyne*, tobacco, resistance, susceptibility, interaction, split-root, temperature.

Root-knot nematodes (*Meloidogyne* spp.) are serious pests of field, vegetable, and horticultural crops (Franklin, 1979; Lamberti, 1979; Sasser, 1979). They are considered the major pest of tobacco throughout the world with an estimated annual yield loss of approximately 15 % (Schneider, 1991). *Meloidogyne arenaria* and *M. incognita*, often in mixed populations, are responsible for substantial flue-cured tobacco yield losses in the southeastern U.S. (Fortnum *et al.*, 1984; Barker, 1989; Gooden *et al.*, 1991). Management systems typically include the use of nematicides, cultural practices, and host-resistance (Todd, 1981; Schneider, 1991). Current resistant cultivars are derivatives of the tobacco cultivar NC 95 and confer protection only against *M. incognita* races 1 and 3. Eisenback (1983), Tedford (1986), Tedford *et al.* (1986), and Ibrahim (1987) indicated that resistance to *M. incognita* failed when tobacco plants were also infected by *M. arenaria*. Eisenback (1983) and Ibrahim (1987) attributed the loss of resistance to a systemic host reaction observable in split-root tobacco plants. Tedford (1986) and Tedford *et al.* (1986) observed this phenomenon under high infection pressure of *M. arenaria* in field experiments.

Host plant resistance has been widely used to suppress *M. incognita* in many southeastern tobacco producing areas. Surveys of root-knot nematode infested fields revealed that almost 60 % of detected *Meloidogyne* populations in South Carolina's tobacco production area were mixed *M. arenaria*/*M. incognita* populations (Fortnum *et al.*, 1984). Cultivation of susceptible tobacco cultivars in fields with mixed infestations of *M. arenaria* and *M. incognita* would allow reproduction of both species. On the other hand, the wide spread use of *M. incognita*-resistant tobacco is likely to increase *M. arenaria* proportions in mixed infestations. Large *M. arenaria* proportions, however, may result in failure of *M. incognita*-resistance in tobacco as observed by Tedford *et al.* (1986). Besides leading to increased damage due to parasitism of both species, such a loss of resistance would invalidate projects aimed at deliberate-

+ *Current address : Department of Plant Pathology, University of Georgia, Athens, GA 30602, USA.*

ly shifting *Meloidogyne* species proportions through rotation schemes and choice of crop cultivars.

A series of greenhouse experiments was conducted to verify the loss of *M. incognita*-resistance due to *M. arenaria* infection. Split root experiments with *M. incognita*-resistant tobacco cultivars were utilized for this purpose. The temperature sensitivity of *M. incognita*-resistance was studied to determine whether the temperature threshold for loss of resistance is lowered through infection with *M. arenaria*.

Materials and methods

Split-root experiments

Tobacco (*Nicotiana tabacum* L.) cvs NC 95 and Coker 176 were used. Both cultivars are resistant to *M. incognita* host races 1 and 3, but susceptible to *M. arenaria*. Tobacco seeds were germinated in trays filled with a peat moss/soil mix. At the two-leaf stage, plants were transplanted into 10 cm^2 square pots filled with sterilized river bottom sand. Plants were grown in the greenhouse for 2 to 4 weeks until *ca.* 5 cm stem length was reached. Tobacco plants were then removed from their pots and roots were washed free of sand with water. Plant roots were separated into two portions using three methods. In method 1, the basal portion of the stem was split longitudinally for 2 cm using a scalpel. In method 2, two equally sized root portions were parted without cutting the stem, thereby minimizing influences triggered by physical damage to the plant. For method 3, the root system was divided without cutting into a proximal and distal portion rather than division into two lateral portions as in methods 1 and 2. To incite systemic reactions in separate root portions, presumably extensive signal translocation, both apical and basipetal, is necessary. Method 3 was developed to determine the direction of spread of a putative signal and to reduce the distance between signal source and target. In each method the two separated root portions of each plant were planted into adjacent 13 cm^2 square plastic pots filled with a sandy loam mixture and secured in trays. To prevent desiccation, moist peat moss covered the roots bridging the two pots of plants in method 3. Plants were inoculated with 0, 5, 15, or 30×10^3 nematode eggs per root portion. One root portion of each plant received eggs of *M. arenaria* and the other received eggs of *M. incognita*. Infestation levels of both species were combined in all possible combinations. A total of five split-root greenhouse experiments were conducted; three using method 1 and one each with methods 2 and 3. In one experiment using method 1, eggs of both species were applied simultaneously, whereas in the remaining four tests, treatment with *M. incognita* was delayed for two or three weeks. All tests were designed as randomized blocks with four replications. Cross contamination of neighboring pots was prevented by plastic shields which were slid between the two pots of each plant. All plants were fertilized daily with Peters Professionnal Peat-Lite Special 20-10-20 fertilizer (Grace-Sierra Horticultural Products Co., Milpitas, CA), dispensed through irrigation water.

Temperature and *M. incognita*-resistance

For the experiment on temperature effects on *M. incognita*-resistance, NC 95 tobacco was planted and grown as described. Approximately 40-day-old plants were transplanted into 13 cm^2 square plastic freezer containers without drainage holes. Soil temperatures were controlled through maintenance of pots in Wisconsin water baths at temperature of 25, 28, 31 and 35 °C. Daily temperature fluctuations were less than 0.5 °C. After the initial plant establishment period of 9 days, pots were infested with *M. arenaria*-Govan at either 0 or 10 000 eggs per plant. Three weeks later, *M. incognita* eggs were applied at the same infestation levels in all possible combinations with *M. arenaria*, resulting in a total of four treatments. Four replications of these treatments were randomized in each of the four water baths.

Nematode isolates and inoculation

M. arenaria race 2 isolates Govan and Pelion from Bamberg and Lexington Counties, S. C., respectively, and *M. incognita* race 3 isolate Witcher from Pickens County, S. C., were maintained on tomato (*Lycopersicon esculentum* Mill.) cv. Rutgers in greenhouse pot cultures. Eggs were extracted from 40 to 60 day-old cultures with 0.05 % sodium hypochlorite (Hussey & Barker, 1973). Plants were inoculated with nematodes by pipetting 5 ml aliquots of eggs in 3 cm deep holes in the soil. Control treatments were infested with similarly prepared extracts of uninfected tomato roots. Holes were filled with soil to prevent desiccation of eggs. Viability and infectivity of *M. incognita* inoculum for each test were verified by inoculating susceptible tomato or tobacco plants at the beginning of each test. These plants were maintained for the duration of each test after which root-galling was assessed.

Harvest and assessment

Roots or root portions were harvested 45 to 60 days after the last infestation. Roots were rinsed free of soil and egg masses were stained with Phloxin B (Dickson & Stuble, 1965). Numbers of egg masses and galls were rated on a 0-5 scale : 0 = 0; 1 = 1-3; 2 =3-10; 3 = 10-30; 4 = 30-100; 5 = > 100 egg masses or galls. The species identity of nematode females found on root portions inoculated with *M. incognita* was determined by esterase isozyme analysis (Esbenshade & Triantaphyllou, 1990) using the PhastSystem automated electrophoresis unit (Pharmacia, LKB Biotechnology, Piscataway, NJ) with a modified protocol (R. S. Hussey, unpbl.). Additionally, for each water bath treatment, 96 egg masses were collected and individually stored at – 80 °C in wells of microtiter plates. DNA from egg masses was extracted, dot blotted, and hybridized to ^{32}P-labeled DNA probes

specific to *M. incognita* (Gárate *et al.*, 1991) or *M. arenaria* as described in Baum *et al.* (1994). Experiments were analyzed independently using the General Linear Models Procedure of SAS (SAS Institute Inc., Cary, NC). T-tests were conducted to determine LSDs and to identify significant effects.

Results

Split-root experiments

M. arenaria did not predispose *M. incognita*-resistant split-root tobacco to infection by *M. incognita* when infested simultaneously or successively with both species. Results of the three experiments using plants with roots and lower stem split (method 1) are shown in Table 1. Simultaneous infestation of both species resulted in limited *M. incognita* reproduction in one treatment with the *M. arenaria* Pelion isolate. In four treatments where *M. incognita* was added 2 weeks after *M. arenaria,* very few *M. incognita* infections were detected. Similar observations were made in treatments in which *M. incognita* eggs were added 3 weeks after infestation with *M. arenaria,* when two treatments with slight *M. incognita* infections were found. *M. incognita* application without prior *M. arenaria* infestation also led in one treatment of cv. Coker 176 (experiment 2) to *M. incognita* reproduction. No significant differences between treatments and controls regarding *M. incognita* reproduction were observed ($P = 0.05$).

Eggs mass and galling ratings of the experiment in which root portions were parted without cutting the stem (method 2), are shown in Table 2. Infestation with *M. arenaria* resulted in nematode reproduction which was positively correlated with initial inoculum density. *M. incognita* failed in all treatments to reproduce on resistant plant material and no galls or egg masses were found.

When roots were divided into proximal and distal portions (method 3), all treatments with *M. arenaria* produced egg masses and galls (Table 3). No effects of *M. arenaria* parasitism on *M. incognita* reproduction were evident ($P = 0.05$). *M. incognita* application following *M. arenaria* infestation resulted in one treatment with one egg mass (15 000 *M. incognita* eggs on the proximal root portion, 2 weeks after 15 000 *M. arenaria* eggs on the distal portion). Control infestations of *M. incognita* without prior *M. arenaria* application similarly led in one case to one egg mass, found on a plant infested with 15 000 eggs at the distal root portion.

Temperature and *M. incognita*-resistance

M. arenaria reproduced well at all soil temperatures. Beginning at 28 °C, *M. incognita* started to reproduce on NC 95 tobacco (Table 4). The number of galls and egg masses of *M. incognita* increased at 31 and 35 °C. In treatments with infestations of both species, *M. incognita* was first found at 31 °C, when 34 % of the recovered

Table 1. *Egg mass (EM) and root galling indices (GI) of* Meloidogyne incognita*-resistant split-root tobacco plants with longitudinally cut stems (method 1) and grown in soil infested with eggs of Govan or Pelion isolates of* M. arenaria *(MA) and with eggs of* M. incognita *(MI) in different combinations and time intervals (0, 2, 3 weeks) after* M. arenaria *infestation.*

Treatment ($\times 10^3$)				MA root half		Mi root half	
	MI-weeks after MA			EM	GI		
MA	0	2	3			EM	GI
Experiment 1 (Coker 176 tobacco)							
0	0			0.0 *b*	0.0 *d*	0.0 *a*	0.0 *a*
0	5			0.0 *b*	0.0 *d*	0.0 *a*	0.0 *a*
0	15			0.0 *b*	0.0 *d*	0.0 *a*	0.0 *a*
MA-Govan							
5	0			3.3 *a*	2.3 *c*	0.0 *a*	0.0 *a*
15	0			4.0 *a*	4.0 *ab*	0.0 *a*	0.0 *a*
5	5			3.5 *a*	3.0 *cb*	0.0 *a*	0.0 *a*
15	5			4.0 *a*	4.3 *ab*	0.0 *a*	0.0 *a*
5	15			3.3 *a*	2.5 *c*	0.0 *a*	0.0 *a*
15	15			3.8 *a*	3.3 *ab*	0.0 *a*	0.0 *a*
MA-Pelion							
5	0			5.0 *a*	5.0 *ab*	0.0 *a*	0.0 *a*
15	0			5.0 *a*	4.8 *ab*	0.0 *a*	0.0 *a*
5	5			5.0 *a*	5.0 *a*	1.0 *a*	0.5 *a*
15	5			5.0 *a*	5.0 *a*	0.0 *a*	0.0 *a*
5	15			5.0 *a*	4.5 *b*	0.3 *a*	0.0 *a*
15	15			5.0 *a*	5.0 *a*	0.0 *a*	0.0 *a*
Experiment 2 (MA-Govan) NC 95 tobacco							
0		0		0.0 *c*	0.0 *c*	0.0 *a*	0.0 *a*
0		5		0.0 *c*	0.0 *c*	0.0 *a*	0.0 *a*
0		15		0.0 *c*	0.0 *c*	0.0 *a*	0.0 *a*
5		0		4.3 *b*	4.8 *b*	0.0 *a*	0.0 *a*
15		0		5.0 *a*	5.0 *a*	0.0 *a*	0.0 *a*
5		5		3.8 *b*	4.0 *b*	0.0 *a*	0.0 *a*
5		15		3.8 *b*	3.5 *b*	0.3 *a*	0.3 *a*
15		5		5.0 *a*	4.8 *a*	0.8 *a*	0.8 *a*
15		15		5.0 *a*	5.0 *a*	0.0 *a*	0.0 *a*
0			5	0.0 *c*	0.0 *c*	0.0 *a*	0.0 *a*
0			15	0.0 *c*	0.0 *c*	0.0 *a*	0.0 *a*
5			5	4.0 *b*	4.0 *b*	0.3 *a*	0.3 *a*
5			15	4.3 *b*	4.0 *b*	0.0 *a*	0.0 *a*
15			5	5.0 *a*	5.0 *a*	0.0 *a*	0.0 *a*
15			15	5.0 *a*	5.0 *a*	0.0 *a*	0.0 *a*
Coker 176 tobacco							
0			0	0.0 *b*	0.0 *b*	0.0 *b*	0.0 *b*
0			15	0.0 *b*	0.0 *b*	0.5 *a*	0.5 *a*
15			0	5.0 *a*	5.0 *a*	0.0 *b*	0.0 *b*
15			15	5.0 *a*	5.0 *a*	0.3 *b*	0.3 *b*
Experiment 3 (MA-Govan) NC 95 tobacco							
0		0		0.0 *d*	0.0 *c*	0.0 *a*	0.0 *a*
0		15		0.0 *d*	0.0 *c*	0.0 *a*	0.0 *a*
0		30		0.0 *d*	0.0 *c*	0.0 *a*	0.0 *a*
15		0		3.8 *c*	4.5 *a*	0.0 *a*	0.0 *a*
30		0		4.5 *ab*	4.8 *a*	0.0 *a*	0.0 *a*
15		15		4.0 *cb*	3.5 *b*	0.0 *a*	0.0 *a*
15		30		4.0 *cb*	4.0 *ab*	0.0 *a*	0.0 *a*
30		15		4.5 *ab*	4.3 *ab*	0.3 *a*	0.0 *a*
30		30		4.8 *a*	4.8 *a*	0.3 *a*	0.0 *a*
0			30	0.0 *d*	0.0 *c*	0.0 *a*	0.0 *a*
30			30	4.8 *a*	5.0 *a*	0.0 *a*	0.0 *a*
Coker 176 tobacco							
0			0	0.0 *b*	0.0 *b*	0.0 *a*	0.0 *a*
0			30	0.0 *b*	0.0 *b*	0.0 *a*	0.0 *a*
30			0	3.8 *a*	3.5 *a*	0.0 *a*	0.0 *a*
30			30	4.3 *a*	4.3 *a*	0.0 *a*	0.0 *a*

Within columns, parameter means with the same letter are not significantly different ($P = 0.05$). Statistical analyses were conducted independently for all three experiments. Within experiment 1, analyses were independent for Govan and Pelion, and within experiments 2 and 3, analyses were independent for NC 95 and Coker 176. Viability of *M. incognita* inoculum was verified by inoculating susceptible tomato or tobacco plants.

Table 2. *Egg mass (EM) and root galling indices (GI) of split-root* Meloidogyne incognita*-resistant NC 95 tobacco plants with roots parted without cutting (method 2) and grown in soil infested with eggs of* M. arenaria*-Govan (MA) and 2 weeks later with eggs of* M. incognita *(MI) in different combinations.*

Treatment ($\times 10^3$)		MA root half		MI root half	
MA	MI	EM	GI	EM	GI
0	0	0.0 *c*	0.0 *c*	0.0 *a*	0.0 *a*
0	*15*	*0.0 c*	0.0 *c*	0.0 *a*	0.0 *a*
0	*30*	*0.0 c*	0.0 *c*	0.0 *a*	0.0 *a*
15	*0*	*3.8 b*	3.3 *b*	0.0 *a*	0.0 *a*
30	*0*	*4.3 b*	3.5 *b*	0.0 *a*	0.0 *a*
15	*15*	*4.3 c*	4.0 *a*	0.0 *a*	0.0 *a*
15	*30*	*4.3 b*	4.0 *ab*	0.0 *a*	0.0 *a*
30	*15*	*4.0 b*	4.0 *ab*	0.0 *a*	0.0 *a*
30	*30*	*5.0 a*	4.8 *a*	0.0 *a*	0.0 *a*

Within columns, parameter means with the same letter are not significantly different ($P = 0.05$). Viability of *M. incognita* inoculum was verified by inoculating susceptible tomato or tobacco plants.

egg masses were *M. incognita* (Table 5). Equal proportions of both species were found in mixed inoculations at 35 °C. No *M. incognita* infections were detected in mixed infestations below 31 °C.

Discussion

Infection with neither *M. arenaria* isolate did affect *M. incognita*-resistance in split-root tobacco plants. Even when present 2 or 3 weeks prior to *M. incognita*, *M. arenaria* was without apparent influence on plant defense against *M. incognita*. Data from the temperature water bath experiment indicated that *M. arenaria* did not influence the temperature sensitivity of *M. incognita*-resistance. *M. incognita* started to reproduce between 28 and 31 °C, regardless of the presence or absence of *M. arenaria*. Slana (1978) similarly found that *M. incognita*-resistance in NC 95 became ineffective at soil temperatures above 30 °C.

Our split-root experiments show that *M. arenaria* parasitism did not elicit a systemic host reaction inducing detectable susceptibility to *M. incognita*. Moreover, mixed *M. arenaria*/*M. incognita* infestations of resistant tobacco with intact roots (water bath experiment) did not result in a loss of resistance to *M. incognita* either. Therefore, *M. incognita*-resistance is stable also in the immediate vicinity of *M. arenaria* infections.

Use of *M. incognita*-resistant tobacco appears appropriate for control of *M. incognita* race 3 even in fields where this race of *M. incognita* coexists with *M. arena-*

Table 3. *Egg mass (EM) and root galling indices (GI) of* Meloidogyne incognita*-resistant NC 95 tobacco plants with roots divided into proximal (top) and distal (bot.) root portions (method 3) and grown in soil infested with eggs of* M. arenaria*-Govan (MA) and with eggs of* M. incognita *(MI) in different combinations and time intervals (2 or 3 weeks) after* M. arenaria *infestation.*

Treatment ($\times 10^3$)						Ma root half		MI root half	
MA		MI (2 weeks)		MI (3 weeks)		EM	GI	EM	GI
top	bot.	top	bot.	top	bot.				
0	0	0	0	0	0	0.0 *f*	0.0 *e*	0.0 *b*	0.0 *a*
	15	0				4.0 *abc*	2.6 *bcd*	0.0 *b*	0.0 *a*
15			0			4.5 *a*	4.3 *a*	0.0 *b*	0.0 *a*
	30	0				2.6 *de*	1.6 *d*	0.0 *b*	0.0 *a*
30			0			2.3 *e*	2.5 *cd*	0.0 *b*	0.0 *a*
	0	15				0.0 *f*	0.0 *e*	0.3 *a*	0.0 *a*
0			15			0.0 *f*	0.0 *e*	0.3 *a*	0.0 *a*
	15	15				3.3 *cd*	2.3 *cd*	0.3 *a*	0.0 *a*
15			15			4.5 *a*	4.8 *a*	0.0 *b*	0.0 *a*
	30	15				4.3 *ab*	4.0 *ab*	0.0 *b*	0.0 *a*
30			15			4.5 *a*	4.5 *a*	0.0 *b*	0.0 *a*
	0			15		0.0 *f*	0.0 *e*	0.0 *b*	0.0 *a*
15					15	4.3 *ab*	4.3 *a*	0.0 *b*	0.0 *a*
	30			15		4.0 *abc*	3.5 *abc*	0.0 *b*	0.0 *a*
30					30	4.5 *a*	4.5 *a*	0.0 *b*	0.0 *a*

Within columns, parameter means with the same letter are not significantly different ($P = 0.05$). Viability of *M. incognita* inoculum was verified by inoculating susceptible tomato or tobacco plants.

Table 4. *Egg mass (EM) and root galling (GI) indices of* Meloidogyne incognita*-resistant tobacco plants grown at controlled soil temperatures and infested with eggs of* M. arenaria *(MA) and 3 weeks later with eggs of* M. incognita *(MI) in different combinations.*

Treatment ($\times 10^3$)			EM	GI
Temperature	MA	MI		
25 °C	0	0	0.0 *c*	0.0 *b*
	10	0	4.0 *b*	3.8 *a*
	0	10	0.0 *c*	0.0 *b*
	10	10	4.5 *a*	3.8 *a*
28 °C	0	0	0.0 *b*	0.0 *b*
	10	0	3.8 *a*	3.8 *a*
	0	10	1.3 *b*	1.0 *b*
	10	10	4.5 *a*	4.0 *a*
31 °C	0	0	0.0 *c*	0.0 *b*
	10	0	3.0 *b*	2.8 *a*
	0	10	4.5 *a*	3.0 *a*
	10	10	4.0 *ab*	3.0 *a*
35 °C	0	0	0.0 *b*	0.0 *b*
	10	0	4.3 *a*	3.0 *a*
	0	10	3.8 *a*	2.8 *a*
	10	10	4.5 *a*	3.3 *a*

Within columns of each temperature, parameter means with the same letter are not significantly different ($P = 0.05$).

Table 5. *Species proportions of* Meloidogyne arenaria *(MA) and* M. incognita *(MI) as determined by DNA hybridization assays in* M. incognita*-resistant NC 95 tobacco after infestation with 10 000 eggs of each species and incubation at controlled soil temperatures.*

Temperature	MA	MI
25 °C	100 % *a*	0 % *b*
28 °C	100 % *a*	0 % *b*
31 °C	66 % *ab*	34 % *ab*
35 °C	53 % *a*	47 % *a*

Within columns, parameter means with the same letter are not significantly different ($P = 0.05$).

ria. Observations made by Eisenback (1983), Tedford (1986), Tedford *et al.* (1986), and Ibrahim (1987) that *M. arenaria* is capable of inducing susceptibility to *M. incognita* are not confirmed by our experiments with the selected nematode isolates.

M. arenaria isolates differ widely in their capability to incite damage and elicit physiological changes in a given host (Carpenter & Lewis, 1991; Noe, 1992; Ibrahim & Lewis, 1993). Other *M. arenaria* isolates may have behaved differently in these experiments. In particular, the two known host races of *M. arenaria* may differ in their potential to predispose resistant tobacco cultivars to *M. incognita* infection, although Ibrahim (1987) observed a loss of *M. incognita*-resistance with a *M. arenaria* race 2 isolate, i.e., the same race we used in our experiments. On the other hand, different *M. incognita* isolates could have benefitted from physiological changes elicited by *M. arenaria*. Eisenback (1983) and Ibrahim (1987) both used race 1 *M. incognita* isolates for their studies, whereas we used race 3. However, *M. arenaria* race 2 and *M. incognita* race 3, as used in our experiments, are the predominant *Meloidogyne* species and races in South Carolina.

Certain management practices aim at shifting mixed root-knot nematode populations away from *M. arenaria* (Fortnum & Currin, 1993) since *M. incognita* races 1 and 3 can be controlled by resistant cultivars and appear to be more susceptible to some nematicides (Barker *et al.*, 1981; Nordmeyer & Dickson, 1985). Such strategies are useful in the design of long term rotation schemes to control nematode infestations in a sustainable agriculture framework. This approach would be jeopardized in case of loss of *M. incognita*-resistance in fields with mixed *M. arenaria*/*M. incognita* infestations. Judging from the presented results, this concern may be unfounded. If induced susceptibility occurs, the frequency is so low that it is hard to detect in the mass of *M. arenaria* reproduction.

Increased incidences of mixed *Meloidogyne* populations in the southeastern U.S. flue-cured tobacco producing area mandate the development of new concepts for root-knot nematode management. Knowledge of peculiarities of mixed infestations, particularly the influence of resistant *vs* susceptible crops in rotations and the effects of nematicidal treatments, is of prime importance for managing polyspecific *Meloidogyne* infestations.

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