Host Resistance and Metam Sodium for Managing Root-knot Nematodes in a Pepper–Cucumber Rotation

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Abstract. Root-knot nematode-resistant ‘Charleston Belle’ bell pepper (*Capsicum annuum* L. var. *annuum*) and metam sodium treatment were evaluated for managing the southern root-knot nematode [*Meloidogyne incognita* (Chitwood) Kofoid and White] in fall-cropped cucumber (*Cucumis sativus* L.). ‘Charleston Belle’ and its susceptible recurrent parent, ‘Keystone Resistant Giant’, were planted as spring crops at Blackville, S.C., and Tifton, Ga. ‘Charleston Belle’ exhibited high resistance and ‘Keystone Resistant Giant’ was susceptible at both locations. After termination of the bell pepper crop, one-half of the plots were treated with metam sodium delivered through the drip irrigation system. Cucumber yields and numbers of fruit were highest for cucumber grown in plots treated with metam sodium following either ‘Charleston Belle’ or ‘Keystone Resistant Giant’; however, root gall severity and numbers of *M. incognita* eggs in the roots were lowest for cucumber grown in plots treated with metam sodium following ‘Charleston Belle’. Conversely, root gall severity and nematode reproduction were highest for cucumber grown in plots following ‘Keystone Resistant Giant’ without metam sodium treatment. Application of metam sodium through the drip irrigation system following a spring crop of root-knot nematode-resistant bell pepper should reduce severity of root gall and reproduction of *M. incognita* as well as increase fruit yield of fall-cropped cucumber.

The southern root-knot nematode, *Meloidogyne incognita* (Chitwood) Kofoid and White, greatly reduces pepper and cucumber yields in the U.S. (Colyer et al., 1998; Thies et al., 1998; Webster et al., 2001). Preplant soil fumigation with methyl bromide is the primary method for controlling root-knot nematodes in bell pepper and cucumber (USDA, 1998). However, production and use of methyl bro-

mid is being phased out in the U.S. (U.S. EPA, 2000). Thus, there is significant interest in developing alternatives to methyl bromide for managing root-knot nematodes in vegetable crops. The use of resistant cultivars as rotational crops for managing root-knot nematodes in subsequent susceptible vegetable crops has been explored by several researchers (Colyer, 1998; Hanna, 2000; Thies et al., 1998; Thies et al., 2004; Webster et al., 2001). Yields of root-knot susceptible bell peppers were heavier and nematode reproduction was less when grown after the root-knot resistant ‘Carolina Cayenne’ pepper than after a susceptible cayenne pepper line (Thies et al., 1998). Melons double-cropped after root-knot resistant tomato exhibited reduced root-galling and produced higher fruit yields than melons grown after susceptible tomato (Hanna, 2000). Cucumber double-cropped after root-knot nematode re-

sistant tomato produced higher yields, and had reduced root gall and lower densities of *M. incognita* second-stage juveniles (J2) in the soil than cucumber grown after susceptible tomato (Colyer et al., 1998; Hanna et al., 1994).

The root-knot nematode-resistant ‘Charleston Belle’ bell pepper was developed and released by USDA–ARS scientists in Charleston, S.C. (Fery et al., 1998). Root-knot nematode resistance in ‘Charleston Belle’ is conditioned by the single dominant *N* gene. In Georgia, ‘Charleston Belle’ and the susceptible bell pepper ‘Camelot’ were planted as a spring crop in rotation with a fall crop of ‘Dixie’ yellow squash (Webster et al., 2001). Soil densities of *M. incognita* J2 observed in ‘Charleston Belle’ plots were about 11% of those observed in ‘Camelot’ plots. Studies in South Carolina and Georgia demonstrated that ‘Charleston Belle’ reduced root gall and increased fruit yields of cucumber grown as a fall crop after ‘Charleston Belle’ compared with cucumber grown after the susceptible ‘Keystone Resistant Giant’ bell pepper (Thies et al., 2004). Thies et al. (2004) suggested that although root-knot nematode resistant bell pepper reduced root-knot nematode damage and increased fruit yields in the subsequent cucumber crop, application of a fumigant nematicide after the resistant pepper spring crop may further reduce root-knot nematode damage and increase yield in the fall cucumber crop. The objective of these studies was to determine the potential of using a resistant bell pepper cultivar in combination with metam sodium for managing *M. incognita* in double-cropped cucumber in South Carolina and Georgia.

Materials and Methods

Field studies were conducted at the Clemson University Edisto Research and Education Center, Blackville, S.C., and the USDA–ARS Crop Protection and Management Research Unit, Tifton, Ga. A spring bell pepper crop and a fall cucumber crop were planted on the same plots at both field sites. One-half of the plots were treated with metam sodium applied through the drip system at termination of the pepper crop. A pair of near-isogenic pepper cultivars that differ for resistance to root-knot nematode conditioned by the *N* gene was used in these studies. The bell pepper cultivars were ‘Charleston Belle’ (*NN*) and ‘Keystone Resistant Giant’ (*nn*) and the cucumber cultivar was ‘Stonewall’.

Egg inocula. *Meloidogyne incognita* race 3 egg inocula was maintained on tomato (*Lycopersicon esculentum* Mill. ‘Rutgers’) in separate greenhouse benches. Egg inocula of *M. incognita* were extracted from infected tomato and bean (*Phaseolus vulgaris* L.) roots using 0.5% NaOCl (Hussey and Barker, 1973).

Blackville bell pepper–cucumber rotation. Pepper seeds were planted in the greenhouse on 13 Mar. 2003. On 8 Apr. 2003, the pepper seedlings were inoculated with about 3,000 *M. incognita* race 3 eggs per seedling. On 22 Apr. 2003, the beds were prepared and covered with 3-mL black plastic. On 23 Apr. 2003, eight cores of soil 20 cm deep were collected

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from each plot and combined into a single sample for nematode extractions. On the same day, pepper seedlings were transplanted into single-row plots on beds 2 m apart. Each plot contained 25 plants spaced 30-cm apart. The experimental design was a randomized complete block with two pepper cultivars $\times$ two metam sodium treatments (an untreated control and 467 L·ha⁻¹) and ten replications. Second-stage juveniles (J2) were extracted from 113 cm³ soil from each sample using the centrifugal flotation method (Jenkins, 1964).

Standard cultural and insect control practices were followed and plots were irrigated with a subsurface drip system (N.C. Vegetable Growers Association, 2004). Soil temperatures were recorded at 60-minute intervals at 5- and 15-cm depths in the row from 6 May through 10 Nov. 2003. Fruit were harvested from 10 plants in the center of each plot on 18 June, 2 July, 10 July, and 16 July 2003. On 16 July 2003, eight pepper root systems with the surrounding rhizosphere soil were dug from each plot. The roots were washed and rated for gall severity using a scale of 1 to 5, where 1 = 0 to 3% root system galled or covered with egg masses; 2 = 4% to 25%, 3 = 26% to 50%, 4 = 51% to 79%, and 5 = $\geq$ 80% root system galled (Thies et al., 1998). *Meloidogyne incognita* eggs were extracted from a 20-g subsample of roots from each plot using 1% NaOCl (Hussey and Barker, 1973). Second-stage juveniles were extracted from 113 cm³ rhizosphere soil as previously described. On 14 Aug. 2003, one-half of the plots were treated with metam sodium at 467 L·ha⁻¹ applied through the drip irrigation system. The black plastic was sprayed with whitewash.

On 4 Sept. 2003, two seeds of 'Stonewall' cucumber were sown in the hole where each bell pepper plant had previously grown. Cucumber seedlings were thinned to one per hill after emergence. Standard cultural and pest control practices were followed and plots were subsurface drip irrigated. Five cucumber plants were dug from the center of each plot on 9 Oct. 2003. The shoots were cut at the base of each plant and weights were recorded. The roots were washed and rated for gall severity and egg mass production using a scale of 1 to 5, as previously described. *Meloidogyne incognita* eggs were extracted from the roots using 1% NaOCl (Hussey and Barker, 1973). Fruit were harvested from five plants in each plot on 6 Nov. 2003.

Tifton bell pepper–cucumber rotation. Field preparation, pest control and cultural practices, and experimental design were as described for the Blackville study. Pepper seedlings were transplanted to the field on 7 May 2003. Soil samples were collected on 16 May 2003 and *M. incognita* J2 were extracted as previously described. Pepper fruit were harvested from all plants in each plot on 8 and 22 July 2003. The root systems of six pepper plants were dug from the center of each plot on 23 July 2003. The roots were rated for gall severity and eggs were extracted from 20 g of fibrous roots as previously described. On 28 July 2003, one-half the plots were treated with metam sodium at 467 L·ha⁻¹ applied through the drip irrigation

system. The black plastic was sprayed with whitewash.

Seeds of 'Stonewall' cucumber were sown on 12 Aug. 2003. Fruit were harvested from all plants in each plot on 14 Oct., 17 Oct., and 20 Oct. 2003. On 27 Oct. 2003, the root systems of all cucumber plants were dug and rated for gall severity. *Meloidogyne incognita* eggs were extracted from the root samples as previously described.

Data analysis. Nematode egg and J2 data were log₁₀(x + 1) transformed to normalize the error variance before analysis (Noe, 1985). Data were analyzed using the GLM procedure of SAS for Windows System Version 6.12 (SAS Institute, Cary, NC). Means were separated using F test and Duncan's multiple range test.

Results and Discussion

Blackville bell pepper–cucumber studies. The soil density of *M. incognita* J2 was about 21

per 100 cm³ soil at planting. Soil temperatures ranged from 12.2 to 39.2 °C at a 5-cm depth and from 15.2 to 37.9 °C at a 15-cm depth from 6 May to 10 Nov. 2003. 'Charleston Belle' had a root gall severity index of 1.0 and 8 *M. incognita* eggs/g fresh root when the study was terminated (Table 1). 'Keystone Resistant Giant' had a root gall severity index of 4.3 and 1,868 *M. incognita* eggs/g fresh root. 'Charleston Belle' had 99.6% fewer ($P \leq 0.0001$) eggs per g fresh root than its susceptible recurrent parent 'Keystone Resistant Giant'. Soil densities of *M. incognita* J2 were 99.4% lower ($P \leq 0.0001$) in plots where 'Charleston Belle' was planted than in plots where 'Keystone Resistant Giant' had been planted. Numbers of fruit produced by 'Charleston Belle' were 18% greater ($P \leq 0.0001$) than numbers of fruit produced by 'Keystone Resistant Giant'.

Cucumber plants grown in plots following the resistant 'Charleston Belle' had less severe root galling ($P \leq 0.05$) and supported fewer (P

Table 1. Gall index, numbers of *Meloidogyne incognita* eggs per g fresh root and second stage juveniles (J2s) per 100 cm³ soil, total fruit yield, and total fruit numbers of 'Charleston Belle' and 'Keystone Resistant Giant' bell peppers.

Pepper cultivar ^a	Gall index ^b	Eggs/g fresh root ^c	J2s/100 cm ³	Yield (kg/plot) ^w	No. fruit/plot ^w
Blackville, S.C.					
Charleston Belle	1.0****	8****	1****	18.7	70****
Keystone Resistant Giant	4.3	1,868	168	16.9	57
Tifton, Ga.					
Charleston Belle	1.3***	1,179***	16****	46.2**	132**
Keystone Resistant Giant	2.0	5,583	184	27.0	84
Combined analysis of both tests					
Charleston Belle	1.2****	98****	5****	32.5**	101***
Keystone Resistant Giant	3.1	3,230	176	21.9	71

^a'Charleston Belle' resistant and 'Keystone Resistant Giant' susceptible to *M. incognita*.

^bGall index: 1 = 0% to 3% root system galled, 2 = 4% to 25%, 3 = 26% to 50%, 4 = 51% to 79%, and 5 = 80% to 100% root system galled.

^cData were log₁₀(x + 1) transformed before analysis. Back transformed data are shown.

^wTwenty-five plants per plot.

****,***,***Significant at $P \leq 0.01$, 0.001, or 0.0001, respectively, according to F tests.

Table 2. Gall index, numbers of *Meloidogyne incognita* eggs per gram fresh root, fresh shoot weight, total fruit yield, and total fruit numbers of 'Stonewall' cucumber double-cropped after 'Charleston Belle' and 'Keystone Resistant Giant' bell peppers with metam sodium application at termination of the bell pepper crop.

Previous pepper cultivar ^a	Metam sodium ^b	Gall index ^c	Eggs/g fresh root ^w	Fresh shoot wt (g/plot) ^v	Yield (kg/plot) ^u	No. fruit/plot ^u
Blackville, S.C.						
Charleston Belle	0	1.3 a ^t	337 b	481 b	4.8 a	9 ab
	+	1.2 a	41 a	512 b	5.5 a	11 b
Keystone Resistant Giant	0	4.6 c	68,140 c	137 a	2.0 a	3 a
	+	3.4 b	23,217 c	453 b	7.7 a	12 b
Tifton, Ga.						
Charleston Belle	0	4.7 ab	9,741 a	---	80.2 a	89 a
	+	4.3 a	11,397 a	---	181.1 b	182 b
Keystone Resistant Giant	0	4.9 b	9,656 a	---	58.4 a	63 a
	+	4.3 a	12,337 a	---	176.7 b	183 b
Combined analysis of both tests						
Charleston Belle	0	3.0 b	2,000 b	---	42.6 a	49 ab
	+	2.7 a	460 a	---	93.3 b	96 bc
Keystone Resistant Giant	0	4.7 d	29,492 c	---	30.2 a	33 a
	+	3.9 c	16,611 c	---	92.2 b	97 c

^a'Charleston Belle' resistant and Keystone Resistant Giant susceptible to *M. incognita*.

^bTreatment with metam sodium at 467 L·ha⁻¹ at termination of pepper study.

^cGall index: 1 = 0% to 3% root system galled, 2 = 4% to 25%, 3 = 26% to 50%, 4 = 51% to 79%, and 5 = 80% to 100% root system galled.

^vData were log₁₀(x + 1) transformed before analysis. Back transformed data are shown.

^wFive plants per plot.

^uTwenty-five plants per plot.

^tValues in a column followed by the same letter are not significantly different according to Duncan's multiple range test ($P \leq 0.05$).

≤ 0.05) *M. incognita* eggs per gram fresh root than plants following the susceptible 'Keystone Resistant Giant' (Table 2). Root galling of cucumber plants grown following 'Keystone Resistant Giant' was reduced in plots that had been treated with metam sodium compared with untreated plots; however, differences were not detected between root gall indices of cucumber plants grown after 'Charleston Belle' in the untreated and metam sodium treated plots. Numbers of eggs per gram fresh root were less ($P \leq 0.05$) in cucumber grown after 'Charleston Belle' than in that grown after 'Keystone Resistant Giant' in both metam sodium treated and untreated plots. Although the numbers of eggs per gram fresh root in cucumber grown after 'Charleston Belle' were less in metam sodium treated plots than in the untreated plots (41 and 337 eggs/g fresh root, respectively), these relatively small differences are probably not biologically significant. Cucumber plants grown following 'Keystone Resistant Giant' in metam sodium treated plots had lower root gall indices ($P \geq 0.05$) than those following 'Keystone Resistant Giant' in untreated plots, but differences were not detected in numbers of eggs per gram fresh root between the metam sodium treatment and untreated control. Fresh shoot weights of cucumber plants grown following 'Keystone Resistant Giant' in plots not treated with metam sodium were 70% to 73% less ($P \leq 0.05$) than those from any other treatment combinations. There was a similar trend of reduced fruit yield of cucumber grown after 'Keystone Resistant Giant' in untreated control plots compared to the other treatment combinations, but significant differences in fruit yield were not detected among treatments.

Tifton bell pepper-cucumber studies. The soil density of *M. incognita* J2 was about 13 per 100 cm³ soil at planting. 'Charleston Belle' had a root gall severity index of 1.3 and 1,179 *M. incognita* eggs per gram fresh root (Table 1). 'Keystone Resistant Giant' had a root gall severity index of 2.0 and 5,583 *M. incognita* eggs/g fresh root. 'Charleston Belle' had 78.9% fewer ($P \leq 0.001$) eggs per gram fresh root than 'Keystone Resistant Giant'. Soil densities of *M. incognita* J2 were 91.3% lower ($P \leq 0.0001$) in plots where 'Charleston Belle' had been planted than where 'Keystone Resistant Giant' had been planted. 'Charleston Belle' produced 42% ($P \leq 0.01$) heavier fruit yields and 37% greater ($P \leq 0.01$) numbers of fruit than 'Keystone Resistant Giant'.

Cucumber plants grown following 'Keystone Resistant Giant' in metam sodium treated plots had somewhat less severe root galling ($P \leq 0.05$) than cucumber following 'Key-

stone Resistant Giant' in untreated plots (gall indices = 4.3 and 4.9, respectively) (Table 2). Differences were not observed among any of the treatment combinations for numbers of *M. incognita* eggs per gram fresh root of cucumber. Cucumber yields were 56% and 67% heavier ($P \leq 0.05$) in metam sodium treated plots following 'Charleston Belle' and 'Keystone Resistant Giant', respectively, than in untreated control plots for the same cultivars. Plants produced 58% more fruit (averaged over cultivars, $P \leq 0.05$) in metam sodium treated plots than cucumber yields in untreated plots.

Results of the combined analysis over locations were included (Tables 1 and 2) because there were interactions for several variables. For example, 'Keystone Resistant Giant' exhibited a susceptible reaction in the Blackville study for both the gall index (4.3) and numbers of eggs per gram fresh root (1,868). However, 'Keystone Resistant Giant' would be classified as resistant based on the root gall index (2.0) in the Tifton study because a susceptible rating is typically defined as ≥ 3.0 on a 1 to 5 rating scale. In contrast, reproduction of *M. incognita* on 'Keystone Resistant Giant' was high (5,583 eggs/g fresh root) in the Tifton study. In addition, fruit yields of 'Charleston Belle' were significantly heavier than its susceptible near-isogenic recurrent parent 'Keystone Resistant Giant'. Thus, it appears that 'Keystone Resistant Giant' was susceptible in the Tifton study.

In the combined analysis over locations, cucumber fruit yields were greatest ($P \leq 0.05$) plots treated with metam sodium following either 'Charleston Belle' or 'Keystone Resistant Giant'. Thus, treatment with metam sodium provided a significant yield benefit for cucumber regardless of which pepper cultivar was grown as the first crop. Although not statistically significant, yields of cucumber grown in untreated plots following 'Charleston Belle' were 29% greater than yields of cucumber in untreated plots following 'Keystone Resistant Giant'. Cucumber grown in plots following 'Charleston Belle' and treated with metam sodium had the lowest ($P \leq 0.05$) root gall severity and lowest ($P \leq 0.05$) numbers of *M. incognita* in the roots. In contrast, root gall severity and nematode reproduction were highest ($P \leq 0.05$) for cucumber grown in plots following 'Keystone Resistant Giant' without metam sodium treatment. Residual nematode pressure and damage to a subsequently planted susceptible crop would likely be reduced in the resistant pepper-cucumber rotation compared to the susceptible pepper-cucumber rotation. We concluded that application of metam sodium

through the drip irrigation system following a spring crop of root-knot nematode resistant bell pepper such as 'Charleston Belle' should reduce severity of root galling and reproduction of *M. incognita* as well as increase fruit yield of fall-cropped cucumber.

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