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Reaction of vegetables and aromatic plants to *Meloidogyne javanica* and *M. incognita*

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ABSTRACT

For this research we used 15 day-old seedlings which were transplanted to 2 L pots and inoculated with 4,000 nematode eggs plus juveniles (J2). After 60 days, the root systems were removed and the number of galls and eggs evaluated and used to calculate the nematode reproduction factor (RF). The tomato cv. Santa Cruz was used as a susceptible control. The experimental design was completely randomized, with six replications. Averages were compared using the Tukey or Scott-Knott test at 5%. For lettuce, Salad Bowl (Mimosa type), Elizabeth and Elisa (Lisa) and Vera cultivars (crisphead), the number of galls and the RF for *M. javanica* were statistically higher than for the control, whereas, for the other vegetable crops, the highest number of galls and eggs were found in chicory and basil. The highest susceptibility to *M. incognita* was observed in Mimosa lettuce cv. Salad Bowl, chicory cultivars, parsley cv. Graúda Portuguesa and basil. Marjoram exhibited no *M. incognita* galls.

Keywords: vegetables, susceptibility, resistance, root-knot nematodes.

RESUMO

Reação de hortaliças e plantas aromáticas aos nematoides *Meloidogyne javanica* e *M. incognita*

Para avaliar a reação, mudas com 15 dias de idade foram transplantadas para vasos de 2 L de capacidade e inoculadas com 4.000 ovos e eventuais juvenis (J2) dos nematoides. Decorridos 60 dias, os sistemas radiculares foram retirados e avaliados quanto ao número de galhas e ovos, determinando-se o fator de reprodução (FR) dos nematoides nas respectivas plantas. Tomateiro cv. Santa Cruz foi utilizado como testemunha. O experimento foi conduzido em delineamento inteiramente casualizado, com seis repetições, e as médias foram comparadas pelo teste Tukey ou Scott-Knott a 5% de probabilidade. Nas alfaces tipo Mimosa cv. Salad Bowl; nas cultivares do tipo Lisa, Elizabeth e Elisa; e na cultivar Vera (tipo crespa), o número de galhas e o FR de *M. javanica* foi superior ao observado para a testemunha; enquanto para as demais oleráceas, os maiores números de galhas e ovos foram para as cultivares de chicória e para o manjeriço. Maior suscetibilidade a *M. incognita* foi observada para a alface tipo Mimosa cv. Salad Bowl, para as cultivares de chicória, salsa cv. Graúda Portuguesa e manjeriço. Apenas manjerona apresentou número de galhas de *M. incognita* igual a zero.

Palavras-chave: hortaliças, suscetibilidade, resistência, nematóide das galhas.

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Vegetables are usually susceptible to nematode attack and their production in tropical countries depends on the correct management of these pathogens (Sikora & Fernandez, 2005). Among vegetables, lettuce (*Lactuca sativa*) is a highly susceptible crop to infestation by root-knot nematodes, especially *Meloidogyne incognita* and *Meloidogyne javanica* (Sikora & Fernandez, 2005). These pathogens have a high reproduction rate, which results in the accumulation of large quantities of eggs in the soil (Campos *et al.*, 2001).

All the lettuce cultivars assessed in the study by Wilcken *et al.* (2004)

were susceptible to *M. incognita* race 2, including the Lucy Brown and Dallas cultivars, where the reproduction factor was 2.33 and 1.25, respectively. Similar results were recently reported by Fernandes & Kulczynski (2009), who observed susceptibility in the Maravilha de Verão, Verônica and Tainá cultivars to *M. incognita*, with reproduction factors ranging from 1.19 to 5.30.

In addition to lettuce, other vegetables are reported in the literature as susceptible to root-knot nematodes, including brassicaceae, solanaceae, cucurbitaceae and liliaceae (McSorley & Frederick, 1995; Ponte *et al.*, 1996;

Walker, 2002; Brito *et al.*, 2007).

A study carried out in the United States calculated the losses in some vegetables from nematode attack (Koenning *et al.*, 1999). The authors reported damage of 5 to 7% in broccoli, 3 to 5% in lettuce, 1 to 5% in onions, reaching 10% in cauliflower and sweet potato. According to the same authors, nematode management in these crops is complex mainly because resistance genes have not been identified in these vegetables. However, for some species and innumerable cultivars, there is no study regarding their reaction to root-knot nematodes and genotypes

can present variable reaction to the different nematode populations. Thus the objective of the present study was to assess the reaction of vegetables and aromatic plants to the root-knot nematodes, *M. javanica* and *M. incognita*.

MATERIAL AND METHODS

Seeds from different vegetable species were germinated in Plantmax® substrate in 128-well extruded polystyrene trays. The experiment was divided into three stages because of seedling and nematode inoculum availability. Thus in the first experiment assessed the lettuce summercrisp cultivars Isabela, Vera and Vanda; iceberg-type cultivars Lucy Brown, Mauren and Tânia; butterhead, cultivars Elisabeth and Elisa; romaine-type cultivars Branca de Paris and Mirella, and the Mimosa-type cv. Salad Bowl. In the second experiment the following were assessed: watercress (*Nasturtium officinale*), Gigante Redondo and Folha Larga cultivars; parsley (*Petroselinum crispum*), Graúda Portuguesa and Lisa Preferida cultivars; chinese cabbage (*Brassica pekinensis*), cv. Híbrida Resistente; broccoli (*Brassica oleracea* var. Itália), Romanesco and Ramoso Piracicaba de Verão cultivars; cabbage (*Brassica oleracea* var. Capitata), Chato and Coração de Boi cultivars; chives (*Allium schoenoprasum*), Todo Ano Nebuka cultivars; broad-leafed chicory (*Cichorium intybus*), Pão-de-Açúcar and Folha Larga cultivars; chicory (*Cichorium endivia*), Gigante Barbarela and Crespa cultivars; spinach (*Spinacia oleracea*), Japonês and Nova Zelândia cultivars; and rocket (*Eruca sativa*), Folha Larga and Apiciatta Folha Larga cultivars. The following were assessed in the third experiment: garlic chives (*Allium tuberosum*), oregano (*Origanum vulgare*), marjoram (*Origanum majorana*), basil (*Ocimum basilicum*), savoury (*Satureja montana*), fennel (*Foeniculum vulgare*) and common chives (*Allium fistulosum*). In all the experiments, the tomato (*Solanum lycopersicum*) cv. Santa Cruz was used as control, considered susceptibility standard.

Fifteen days after germination, the seedlings were transplanted to 2 liter polyethylene pots, containing a previously autoclaved (2h/120°C) 2:1 (v:v) soil and sand mixture. Three days after transplant, the soil in each pot was infested with approximately 4,000 eggs and occasional second stage juveniles of *M. javanica* or *M. incognita*. To obtain the inoculum the pure nematode populations, kindly donated by the Nematode Laboratory at the Federal University of Viçosa, were multiplied in the tomato cv. Santa Cruz for approximately three months. After this period, the nematodes were extracted from the root system using methodology by Hussey & Barker, adapted by Boneti & Ferraz (1981). The eggs plus juveniles were counted using a Peters chamber and an optical microscope and the suspension obtained was calibrated for 1,000 eggs plus juveniles nematodes in 1 mL water. The infestation was made in four orifices, opened in the soil around the plant and approximately 1 mL placed per orifice.

Sixty days after infestation, the plants were carefully removed from the pots and the root system separated from the canopy. The galls were counted directly in the root system and later the eggs were extracted (Boneti & Ferraz, 1981) and the total number (eggs and second stage juveniles) plotted in the formula $RF = Fp/Ip$, where RF is the reproduction factor, Fp the final population and Ip the initial population (Oostenbrink, 1966).

A complete randomized design was used with six replications for each treatment. The values obtained were submitted to analysis of variance and the means compared by the Tukey test and the Scott-Knott test at the level of 5% probability.

RESULTS AND DISCUSSION

The lettuce cultivars assessed did not present immunity reaction ($RF = 0$) to *M. javanica* and *M. incognita* (Table 1). For the Mimosa-type lettuce cv. Salad Bowl, the number of *M. javanica* galls was statistically superior to that of the control, the tomato cv. Santa Cruz,

with a RF equal to 4.3. Generally, lower RF was observed for the iceberg and romaine-type cultivars. The butterhead-type cv. Elisa and Elizabeth presented $RF > 1$, as for the summercrisp type cv. Vera and Mimosa-type cv. Salad Bowl. The susceptibility of the cv. Elisa to *M. javanica* had been previously reported by Santos (1995) in a study with the lettuce cultivar after the area had been planted with tomato in the field and yield decreased from 17 to 78%. Charchar (1991) also reported that the butterhead cultivars were susceptible to *Meloidogyne* spp.

Regarding *M. incognita*, only the cv. Vera presented a smaller number of galls than that observed in the control treatment (Table 1). Although the cultivars, except for the Mimosa-type cv. Salad Bowl, presented $RF < 1$, the high number of galls showed the susceptibility to nematodes. The results reported here are in line with those presented by Wilcken *et al.* (2004), where all the lettuce cultivars assessed were susceptible to *M. incognita* race 2.

Charchar & Moita (2005) reported the Vitória and Regina lettuce cultivars as highly susceptible to *M. incognita* race 1 and *M. javanica* and as susceptible the Vitória de Verão, Babá, White Boston and Piracicaba-65 cultivars, while Carneiro *et al.* (2000) reported that the summercrisp Crespa Rápida and Livia cultivars presented reactions that ranged from moderately to highly susceptible to the *M. javanica* and *M. incognita* race 3. In addition to *M. javanica* and *M. incognita*, other root-knot nematode species, such as *M. arenaria* and *M. hapla* can damage the lettuce crop (Taylor & Sasser, 1978; Charchar *et al.*, 1999; Carneiro *et al.*, 2000).

The reaction of the cultivars belonging to the same type of lettuce could vary, for example, *M. javanica*, whose reproduction factor in the summercrisp type lettuces ranged from 0.74 to 2.55 (Table 1). Charchar & Moita (2005) worked with mixed populations consisting of *M. javanica* and *M. incognita* race 1 and observed that the reaction of the butterhead type lettuce cultivars varied from moderately resistant to highly susceptible, and the summercrisp lettuce, from highly

Table 1. Galls and eggs numbers and reproduction factor (RF) of *Meloidogyne javanica* and *M. incognita* in different lettuce cultivars (número de galhas, de ovos e fator de reprodução (FR) de *Meloidogyne javanica* e *M. incognita* em diferentes cultivares de alface). Umuarama, UEM, 2009.

Type	Cultivar	<i>M. javanica</i>			<i>M. incognita</i>		
		Galls ¹	Eggs ¹	RF ²	Galls ¹	Eggs ¹	RF ²
Summercrisp	Isabela	83.5 a	2,954.3 a	0.74	227.5 b	1,613.0 a	0.40
	Vera	185.8 ab	10,182.8 ab	2.55	21.5 a	1,085.5 a	0.27
	Vanda	27.8 a	1,873.3 a	0.47			
Iceberg-type	Lucy Brown	10.3 a	748.3 a	0.19			
	Mauren	47.0 a	1,524.3 a	0.38	75.8 ab	3,774.3 a	0.94
	Tânia	34.7 a	506.0 a	0.13			
Butterhead	Elisabeth	83.8 a	5,857.0 ab	1.46	105.3 ab	1,123.3 a	0.28
	Elisa	174.0 ab	14,492.5 ab	3.62	237.3 b	1,545.0 a	0.39
Romaine	Branca Paris	58.8 a	658.3 a	0.16	214.3 ab	2,375.8 a	0.59
	Mirella	17.5 a	1,969.0 a	0.49	49.0 ab	2,440.0 a	0.61
Mimosa	Salad Bowl	672.5 c	17,214.5 bc	4.30	171.3 ab	5,253.7 a	1.31
Control	Tomato	307.8 b	30,744.3 c	7.69	226.5 b	19,291.5 b	4.82

¹Means followed by the same letter do not differ among themselves according to the Scott-Knott test at 5% probability; ²Reproduction factor (¹médias seguidas pela mesma letra não diferem entre si pelo teste Scott-Knott a 5% de probabilidade. ²Fator de reprodução).

resistant to susceptible to the nematodes. However, the authors reported that generally the summercrisp type lettuces were more resistant than the butterhead type. Similar results were also reported by Gomes *et al.* (2000) and Fiorini *et al.* (2005). Some cultivars were studied as resistance sources, such as Grand Rapids and Salinas. Other lettuce cultivars, such as Sea Green Nua, Bix, Romana Balão and Ferry Morse, are reported as resistant or highly resistant to *Meloidogyne* species (Charchar & Moita, 1996).

Lettuce cultivar susceptibility can vary in function of the nematode population, including the *M. incognita* races, and the conditions under which the experiment is carried out, mainly because some cultivars can present field resistance. The cv. Salad Bowl, that was susceptible to *M. javanica* under controlled conditions in the present study, was reported by Charchar & Moita (2005) as highly resistant to *M. incognita* race 1 and *M. javanica* in the field.

In addition to the lettuce cultivars, most of the vegetables studied in the second trial presented high egg numbers and especially high gall numbers, showing the susceptibility to nematode (Table 2). The smaller numbers of galls and eggs of *M. javanica* were

obtained for the parsley cultivars and for the rocket cv. Folha Larga. On the other hand, high number of galls was observed in the broad-leafed chicory and chicory cultivars, and the chinese cabbage cv. Híbrida Resistente, broccoli cv. Romanesco, chive cv. Todo Ano Nebuka and spinach cv. New Zealand. The highest number of *M. javanica* eggs was observed in the chicory cultivars that were equal to those found in the control treatment, the tomato cv. Santa Cruz (Table 2).

The rocket cv. Folha Larga, broad-leaved chicory cv. Folha Larga and chinese cabbage cv. Híbrida Resistente were the vegetables assessed in the second experiment that presented the smallest number of *M. incognita* galls and eggs (Table 2). Similar to that observed for *M. javanica*, the highest number of *M. incognita* eggs was found in the chicory cultivars. The high susceptibility of chicory to *M. incognita* had been observed previously by Walker (2002), who reported that the increase in the initial nematode population reduced plant dry weight and size, but did not mention the cultivars used in the experiment.

Analysis of the brassica reaction showed that generally the highest number of galls and eggs were obtained for *M. javanica*, especially for the

cabbage cv. Chato with RF=1.07 (Table 2). Smaller number of *M. incognita* eggs was recorded in chinese cabbage and in the broccoli Ramoso Piracicaba de Verão and rocket cv. Apiciatta Folha Larga. The susceptibility of these plants to root-knot nematodes was reported by some researchers (McSorley & Frederick, 1995; Almeida *et al.*, 1997; Brito *et al.*, 2007). Of the cultivars of *B. oleracea* assessed by Brito *et al.* (2007) for susceptibility to *Meloidogyne mayaguensis*, currently called *M. enterolobii*, only the cv. Acephala presented resistance reaction, while the others, including broccoli, cabbage and mustard were susceptible to this nematode species with RF ranging from 5.80 to 12.10. Similar results were obtained in the experiments carried out by McSorley & Frederick (1995), in which broccoli, cabbage and Chinese cabbage presented IG>3 (index gall) for *M. javanica* while for *M. incognita* race 1 and race 3, the IG ranged from 1.42 for broccoli to 4.33 for Chinese cabbage and from 3.17 to 4.58 in the same hosts, respectively. In the study, the authors reported that the number of nematodes recovered from the root system varied considerably, a characteristic also observed in the results of the present study (Table 2). In the experiment by Almeida *et al.* (1997), the cabbage cv.

Table 2. Galls and eggs numbers and reproduction factor (RF) of *Meloidogyne javanica* and *M. incognita* in different vegetables (número de galhas e de ovos e fator de reprodução (FR) de *Meloidogyne javanica* e *M. incognita* em diferentes oleráceas). Umuarama, UEM, 2009.

Vegetable	<i>M. javanica</i>			<i>M. incognita</i>		
	Galls ¹	Eggs ¹	RF ²	Galls ¹	Eggs ¹	RF ²
Watercress 'Gigante Redondo'	32.7 a	2,370.0 b	0.59	64.2 b	1,142.8 ab	0.29
Watercress 'Folha Larga'	35.6 a	1,337.2 a	0.33	49.8 b	1,387.0 ab	0.35
Broad-leafed chicory 'Pão-de-açúcar'	527.2 c	2,204.5 b	0.55	51.4 b	326.6 a	0.08
Broad-leafed chicory 'Folha Larga'	451.5 c	224.7 a	0.06	7.3 a	358.5 a	0.09
Spinach 'Japonês'	62.2 a	595.5 a	0.15	171.8 bc	1,173.6 ab	0.29
Spinach 'Nova Zelândia'	320.8 b	3,243.0 bc	0.81	106.0 b	955.2 ab	0.24
Chicory 'Gigante Barbarela'	830.7 c	28,158.7 c	7.04	297.3 c	9,326.7 c	2.33
Chicory 'Crespa'	511.3 c	16,342.3 c	4.09	324.7 c	9,893.0 c	2.47
Rocket 'Apreciatta Folha Larga'	56.0 a	3,337.3 bc	0.83	54.5 b	230.7 a	0.06
Rocket 'Folha Larga'	17.0 a	1,132.2 a	0.28	13.7 a	445.5 a	0.11
Chinese cabbage 'Híbrida Resistente'	237.0 ab	1,718.3 a	0.43	1.0 a	188.7 a	0.05
Broccoli 'Romanesco'	202.8 ab	3,171.3 bc	0.79	31.5 b	260.5 a	0.07
Broccoli 'Ramoso Piracicaba de Verão'	75.5 a	1,689.8 a	0.42	63.7 b	213.7 a	0.05
Cabbage 'Chato'	84.8 a	4,272.0 bc	1.07	72.5 b	499.8 a	0.12
Cabbage 'Coração de Boi'	62.8 a	1,051.5 a	0.26	62.2 b	447.2 a	0.11
Parsley 'Graúda Portuguesa'	12.3 a	1,254.2 a	0.31	98.5 b	4,633.7 bc	1.16
Parsley 'Lisa Preferida'	28.4 a	3,408.0 bc	0.85	166.3 bc	2,043.5 ab	0.51
Chives 'Todo Ano Nebuka'	109.3 ab	2,156.7 b	0.54	43.7 b	2,938.2 b	0.73
Control	355.5 bc	17,932.7 c	4.48	*	*	

¹Means followed by the same letter do not differ among themselves according to the Scott-Knott test at 5% probability; ²Reproduction factor; *Dead plants ('médias seguidas pela mesma letra não diferem entre si pelo teste Scott-Knott a 5% de probabilidade; ²Fator de reprodução; * Morte das plantas).

Table 3. Galls and eggs numbers and reproduction factor (RF) of *Meloidogyne javanica* and *M. incognita* in different aromatic plants (número de galhas e de ovos e fator de reprodução (FR) de *Meloidogyne javanica* e *M. incognita* em diferentes plantas aromáticas). Umuarama, UEM, 2009.

Aromatic plants	<i>M. javanica</i>			<i>M. incognita</i>		
	Galls ¹	Eggs ¹	RF ²	Galls ¹	Eggs ¹	RF ²
Common chives	4.7 a	264.8 a	0.07	50.5 a	764.2 a	0.19
Garlic chives	0.8 a	1,273.5 a	0.32	1.3 a	1,406.3 a	0.35
Basil	314.2 b	9,387.5 ab	2.35	443.7 b	18,986.7 b	4.75
Oregano	3.5 a	286.7 a	0.07	26.2 a	1,878.3 a	0.47
Marjoram	4.7 a	2,672.5 a	0.67	0.0 a	415.2 a	0.10
Fennel	13.2 a	823.0 a	0.21	12.8 a	1,285.8 a	0.32
Savoury	8.8 a	1,463.8 a	0.37	26.5 a	8,313.3 ab	2.08
Control	323.7 b	31,128.7 b	7.78	368.7 b	9,173.2 ab	2.29

¹Means followed by the same letter do not differ among themselves according to the Tukey test at 5% probability; ²Reproduction factor ('médias seguidas pela mesma letra não diferem entre si pelo teste de Tukey a 5% de probabilidade; ²Fator de reprodução).

Coração de Boi was fairly resistant to *Meloidogyne megadora*, with a final number of nematodes equal to zero, but the authors observed the presence of galls in the root system (IG=3).

The reaction to root-knot nematode has been variable in the parsley crop.

For the crops assessed in the present study, the number of *M. javanica* galls ranged from 12.3 to 28.4 and from 98.5 to 166.3 for *M. incognita* and the greatest number of *M. incognita* eggs was found in the parsley cv. Graúda Portuguesa (Table 2). Almeida *et al.*

(1997) observed *M. incognita* and *M. javanica* IG and RF equal to zero in the parsley crop. On the other hand, high susceptibility of parsley to *M. incognita* was observed by Walker (2002) with gall indices equal to those observed in the control treatment, regardless of the

initial nematode population. The authors further observed that the increase in inoculum density reduced the plant dry weight.

The chives species assessed in the second experiment (*A. schoenoprasum*) presented a high number of *M. javanica* and *M. incognita* galls (Table 2). On the other hand, when Walker (2002) compared the number of *M. incognita* galls in *A. schoenoprasum* (0.9) and the control (109.0), low nematode reproduction was observed. Ponte *et al.* (1996) reported shallots (*Allium ascalonicum*) as susceptible to *M. incognita* and *M. javanica*, but did not report details regarding reproduction of these *M. incognita* species in the host.

When the reaction of aromatic plants was assessed, it was observed that common chives had a low number of *M. incognita* and *M. javanica* galls (Table 3), unlike that observed for *A. schoenoprasum* cv. Todo Ano (Table 2). Regarding garlic chives, there are few studies on the reaction of this crop to root-knot nematode. In a study carried out in Korea (Kim & Lee, 2008), *M. incognita* was indicated as one of the causes of garlic chives decline. In the present study, low number of galls and eggs of both the *Meloidogyne* species were observed in this crop (Table 3).

Among the plants assessed in the third experiment, the highest number of *M. javanica* and *M. incognita* galls was observed for basil that was statistically equal to the control treatment (Table 3). These results corroborated those reported by Moreno *et al.* (1992), where basil presented susceptibility reaction to different *Meloidogyne* species and races. The same authors observed resistance reaction in oregano to root-knot nematodes while in the present study the number of galls and eggs was significantly lower for *M. javanica* (Table 3).

Marjoram presented few *M. javanica* galls and no *M. incognita* galls. Similar results were reported by Moreno *et al.* (1992) where a gall index of this plant was zero for all the *Meloidogyne* species studied. The consistency of the results between the two studies was also observed for savoury.

Fennel, that in the present study

presented low number of galls and eggs of both species (Table 3), was susceptible to *M. incognita* (RF=0.32) in the assessment carried out by Park *et al.* (2007). Different nematode populations, including races, genotypes of the plant or experimental conditions can account for the inconsistency in the results.

The present study showed the variation in the reaction of vegetables to different species of root-knot nematodes. This observation refers to some factors that should be considered when it is necessary to manage these pathogens. The first is the importance of studying options to form a crop rotation scheme that should include non-host, resistant and moderately resistant species and also the relevance of knowing the nematode species present in the area. It is emphasized that the data presented here are preliminary and results different from those observed in the present study may be obtained considering variation in the nematode inoculum concentration or in the experimental conditions.

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