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Prospection and production of Solanaceae species resistant to the root knot nematode

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Plant species of Solanaceae family are affected by numerous pathogens worldwide. Among them is the root-knot nematode which hinders the establishment of crops in the field, reducing their production capacity. This work aims to select Solanaceae species tolerant or resistant to root-knot nematode under the climatic conditions of Fortaleza, Ceará. Two experiments were performed. For both, the followings were evaluated: 'Santa Clara' tomato, hybrid 'T92', cherry 'Carolina' and 'Laranja'; gilo 'Comprido Grande Rio'; eggplant 'Comprida Roxa'; Pepper 'Cayenne'; and chili 'All Big'. Each treatment had six replicates. In the first trial, the number of twigs and egg masses per root, the aboveground height and fresh root weight at 60 days after inoculation (DAI) were evaluated. In the second trial, the reproduction factor of the 130 DAI, total number and weight of fruits, as well as productivity were evaluated. The tomato 'Santa Clara', 'Carolina', 'Laranja', the eggplant and gilo had the major infestations of *Meloidogyne incognita*; however, only tomato 'Santa Clara showed decreased productivity among all the cultivars. On the other hand, in the hybrid 'T92' and chili, there was no nematode reproduction, no eggs mass and reproduction factor was zero. Thus, it is concluded that Pepper 'Cayenne', 'All big' Chili 'All Big' and 'T92' hybrid tomato are immune to Meloidogyne *incognita*, so they can be tested in resistant rootstock trials for susceptible commercial tomato plants.

Key words: Root-knot nematode, *Solanum lycopersicum, Solanum melongena, Capsicum annuum, Solanum gilo, Meloidogyne incognita*.

INTRODUCTION

Vegetable production in Brazil has great economic, social and nutritional importance. Due to new technologies and developed cultivation techniques the production of vegetables increased by 33% and their productivity by 38% (Carvalho, 2013). According to the Brazilian Association of Seeds and Seedlings Commerce (Associação Brasileira do Comércio de Sementes e Mudas - ABCSEM, 2014), more than 94 billion reais were

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moved in 2012 in the agricultural chain that involves the vegetable production segment. In total, approximately 20 million tons of -vegetable crops were produced. Socially, the production of this group of plants favored employability in Brazil, since only in 2012, about 2 million people were directly employed in this type of activity (ABCSEM, 2014).

In addition to the above, the production of vegetables promotes human health, by providing a nutritionally diet rich in fiber, vitamins and minerals (Machado, 2008), which are key components to a healthy and balanced diet.

Despite the wide applicability, consumption and productivity of the species of the Solanaceae family, edaphoclimatic factors such as temperature, light, wind, nutrient concentration and phytosanitary problems may adversely affect the establishment of these crops in the field, thus reducing their production capacity and making them more expensive to purchase by consumers. Some of these factors are often addressed through the planning and selection of areas for cultivation. However, issues relating to phytosanitary problems such as the presence of unwanted organisms cannot always be predicted, given the impossibility of having full control of the entrance of a given pathogen in the production area. Among the many organizations that affect the production of tomato, great emphasis can be given to the root-knot nematode (*Meloidogyne* spp). This pathogen belongs to the genus Meloidogyne (Goeldi - 1887), which has four main species: *Meloidogyne incognita*, *Meloidogyne javanica*, *Meloidogyne arenaria* and *Meloidogyne hapla* with widespread distribution throughout the world. It composes the nematode group of greater importance for vegetables (Pimenta and Carneiro, 2005), affecting the vast majority of cultivated solanaceae (Pernezny et al., 2003).

Cultural control methods including crop rotation, fallowing, the use of pesticides, and solarization are often employed in the management of root-knot nematodes. Nevertheless, although such techniques present some efficiency, some have operational and economic drawbacks that make it difficult and often hamper their implementation by producers. For example, the use of solarization limits the production area for crops, which is of particular importance for small farmers that cannot grow in the area being solarized, or cannot solarize its infested area because of the rainy season (Baptista et al., 2007).

Furthermore, the use of chemical pesticides is often associated with negative environmental impacts and increased production costs. The development and deployment of nematode-resistant cultivars is an efficient, cost-effective and environmentally-friendly option for nematode management. In previous reports, the use of host plant resistance decreased nematode reproduction with effectivity comparable to chemical control (Cook and Evans, 1987; Starr and Roberts, 2004).

Given the above, the objective of this study is to identify species and cultivars of the Solanaceae family that have tolerance or resistance characteristics to most frequent worldwide species of root-knot nematode, *M. incognita.*

MATERIALS AND METHODS

For the development of this work two trials were conducted. The work completion period was from September 2014 to February 2015. For both tests, the design was completely randomized in a factorial 2 x 8 (two ground conditions, infested and uninfested with root-knot nematodes, and eight species / cultivars: 1) Tomato 'Santa Clara' from Santa Cruz group (susceptible to root-knot nematode); 2) Hybrid tomato 'T92'; 3) Cherry tomato (*S. lycopersicum* var cerasiforme) 'Carolina'; 4) Cherry tomato 'Laranja'; 5) Gilo (*Solanum gilo* Raddi) 'Comprido Grande Rio'; 6) Eggplant (*Solanum melongena* L.) 'Comprida Roxa'; 7) Chili (*Capsicum annuum*) 'Cayenne'; and 8) Chili 'All Big' (*C. annuum* var annuum L.). One plant was cultivated per pot and each species was replicated six times under each of the two treatments.

The seedlings were grown in polyethylene trays with 162 cells filled with substrate produced from organic compound and vermiculite in the proportion of 9:1. The physical and chemical composition of the soil used for filling the trays is shown in Table 1. After sowing, the trays were allocated in a covered structure with 30% shading, where they remained for about 30 days until they were transplanted to the final location for production. The physical and chemical characteristics of the compound used for filling the trays and pots cultivation of Solanaceae are as follows (Table 1).

When the seedlings had three to four true leaves, they were transplanted to the pots with 8 L capacity, and cultivated until harvest. Fifteen days after transplanting the plant growth was supported by vertical staking according to the protocol by Guimarães et al. (2007, 2008), using polyethylene strings.

Irrigation was carried out twice a day with a system characterized as micro-sprinkler type. Ridge planting was performed 30 days after transplanting. Other cultural practices such as thinning and hoeing were performed as needed following the recommendations of Filgueira (2008).

To obtain the inoculum, the nematode was cultured on the roots of cherry tomato 'Carolina', (*S. lycopersicum* var cerasiforme) grown on site. 60 days after inoculation the infested roots were removed from the soil, carefully washed, wrapped in plastic bags and taken to the Phytopathology Laboratory in Universidade Federal do Ceará. Eggs of the root-knot nematode population were extracted using Coolen and D'Herde's (1972) technique and approximately 5,000 eggs were injected into the soil of each plant at a depth of 3 cm two days after transplanting.

For evaluation of the results, the following parameters were considered: number of galls (NG); galls index (GI); mass number of external eggs (ME); egg mass index (EMI). The GI and EMG in the roots were represented by a scale of 1 to 5, according to Taylon and Sasser (1978), modified by Hadisoeganda and Sasser (1982).

After 130 days of inoculation, total fresh roots were harvested to determine the reproduction factor (RF) and reproduction index (RI) as an estimate of the reproductive capacity of the nematodes: RF = Fp/Ip, where Fp = final nematode population and Ip = initial nematode population. In this parameter, plant species are classified as immune (RF = 0), resistant (RF < 1.0) and susceptible (RF > 1.0) (Oostenbrink, 1966).

The rendering index (RI) was obtained by the ratio between the average number of eggs per gram of root plants of each treatment and the average number of eggs per gram of root tomato 'Santa Clara' (pattern) multiplied by 100 (Taylor, 1967). The classification corresponds to susceptible (S) when RI > 50%, slightly resistant (SR) if RI 26 to 50%, moderately resistant (MR) if RI from 11 to

Table 1. Physical and chemical composition of the soil used for the production of Solanaceae seedlings.

Chemical characteristics	Value	Unit	Puller
pH	6.80	mg dm^{-3}	In H_2O, KCl and $CaCl_2$ – Ratio 1:2.5
P	50.40	mg dm^{-3}	Puller Mehlich 1
K	57.00	$cmol_c$ dm^{-3}	Puller Mehlich 1
Ca^{2+}	20.30	$cmol_c$ dm^{-3}	KCl – 1 mol L^{-1}
Mg^{2+}	6.90	$cmol_c$ dm^{-3}	KCl – 1 mol L^{-1}
Al^{3+}	0.00	$cmol_c$ dm^{-3}	KCl – 1 mol L^{-1}
H+AL	1.49	$cmol_c$ dm^{-3}	Acetate calcium 0.5 mol L^{-1} - pH 7.0
SB	27.35	$cmol_c$ dm^{-3}	Exchangeable basic sum
CTC (t)	27.35	$cmol_c$ dm^{-3}	Effective capacity of cation exchange
CTC (T)	28.84	%	Capacity of cation exchange to the pH 7.0
V	95.00	%	Base saturation index
M	0.00	%	Aluminum saturation index

Table 2. Susceptibility classification of the plants according to the number of root-knot and egg mass (Taylor and Sasser, 1978) modified by Hadisoeganda and Sasser (1982).

Number of root-knot and egg masses	Index number	GI/EMI	Plants classification
0	0	0.0-1.0	Highly resistant
01-02	1	1.1-3.0	Very resistant
03-10	2	3.1-3.5	Moderately resistant
11-30	3	3.6-4.0	Lightly resistant
31-100	4	4.1-5.0	Susceptible
>100	5	-	-

25%, very resistant (VR) if RI from 1 to 10%, highly resistant (HR) if RI < 1.0% and immune (I) when there is no reproduction (Table 2).

To characterize the productivity of Solanaceae species, periodic harvests of fruits were performed, to assess the number of fruits produced and as well as total fruit fresh weights.

Statistical analysis was performed on collected data using the Scott - knott. The obtained data were submitted to analysis of variance by the software Sisvar (Ferreira, 2010) and, once difference between the treatments was observed, they were compared by the Scott-Knott group test's at 5% level of significance.

RESULTS AND DISCUSSION

After the evaluation of the plants used in the determination of susceptibility (Table 2) it can be seen that tomatoes 'Laranja' and 'Carolina'; eggplant 'Comprida Roxa' an gilo 'Comprido Grande Rio' - showed the highest average number of galls and number of egg masses 60 days after inoculation (DAI) as well as the reproduction factor (RF) in the 130 DAI of the plants.

Such results indicate that the susceptibility of the species to *M. incognita*, gilo (S. gilo) IR showed less than 50% indicating a slight resistance (SR) to the nematode (Table 3). The susceptibility of other tomato cultivars to more widespread nematodes in the middle of agricultural production (*M. javanica, M. incognita, M. arenaria* and *M. enterolobii*) has been reported by other researchers in other studies (Talavera et al., 2009; Bitencourt and Silva, 2010).

In comparison with other plant species, hybrid tomato 'T92', the pepper 'All Big and chili 'Cayenne' are considered immune to nematode infection (Table 3) and immune (I) as FR and RI were zero, according to the criteria of Oostenbrink (1966) and Taylor (1967), respectively. This result was expected for the long life hybrid tomato 'T-92' that commercially has, as one of its main features, resistance to nematode galls *M. incognita*, *M. arenaria* and *M. javanica*. The results obtained with the chili 'Cayenne' were similar to those found by other authors for other species within the genus Capsicum. Carneiro et al. (2000) observe two cultivars of sweet chili (*C. annuun*) resistant to *M. javanica* and *M. arenaria*. Recently, Rosa (2013) observed immunity to *M. javanica* in different cultivars of chili.

Pepper cultivar “All Big” showed immunity to root-knot nematode infection which was comparable to previous studies involving a variety of *Capsicum* spp. (Carnerio et al., 2000). In general, what could be seen from the

Table 3. Evaluation of parameters: number of galls, egg mass, number of eggs and reproduction factor of *Meloidogyne incognita*.

Species/Cultivars	Number of root-knot[1,2]	GI	Number of egg mass	EMI	Number of eggs[1]	Reproduction factor[1]	RI
Tomato 'Laranja'	495	5	77.67	4	250,667	50.13	89
Tomato 'Carolina'	500	5	75.83	4	444,000	88.80	96
Tomato 'T92'	0	0	0.00	0	0	0	0
Chilli 'All Big'	0	0	0.00	0	0	0	0
Pepper 'Cayenne'	1	1	0.00	0	0	0	0
Eggplant ‘Comprida Roxa '	500	5	78.50	4	1,538,667	307	145
Gilo ‘Comprido Grande Rio’	500	5	72.00	4	464,000	93	47
Tomato ‘Santa Clara’	500	5	72.83	4	535,000	107	100

[1]Average of six replicates; accounted for up to a maximum of 500 root-knot. GI = Galls index of root-knot nematode; EMI = Egg mass index; RI = reproduction index.

various research results already published and the information generated from this study is that the genetic diversity among different cultivars of pepper and chili available can be considered the main factor responsible for resistance or susceptibility of plants to the species of the root-knot nematodes, since there are wide varieties that exhibit resistance to more than one kind of nematode; simple resistance, being resistant to one kind of nematode; and complete absence of resistance (susceptibility).

For the observed characteristic of the shoot height, no difference was observed between the plants grown under different conditions of soil infestation for cultivars of tomato cherry group, hybrid tomato, pepper, chili and eggplant. However, for tomato 'Santa Clara' and gilo, differences were found between the plants grown in soil infested with the pathogen or not. For the tomato 'Santa Clara' there was greater shoot height for plants grown in soil without infestation, while gilo (*S. gilo*) showed higher shoot values for plants grown in an environment infested by the pathogen (Table 4).

With the exception of plant cultivars “T92”, “Comprido Grande Rio” and “Comprida Roxa”, all species demonstrated increases in root mass during cultivation in nematode-infested soil. For the characteristic evaluated root mass, except for the tomato hybrid 'T92' and eggplant's cultivar that had lower average values for plants grown in an environment infested with nematodes, and the gilo's cultivar (*S. gilo*), which showed no significant difference in mass between plants grown in soil infested by nematode or not, all other cultivars of each evaluated species produced higher average of root mass when grown in infested soil with the study pathogen (Table 5).

In contrast to Sharma et al. (2005), increased shoot height and root mass was observed in pepper variety “Cayenne”. The highest fresh weight of roots observed, generally, in infested treatments is related mainly with the thickening of galls on nematodes infestation, which causes certain increase in mass at the site of infestation. The galls on each root system showed that this accumulation contributes to a significant increase in fresh root mass compared to uninfested plants.

For production characteristics, for all other species and cultivars no differences in yield were observed between the two soils used for cultivation, both infested and non-infested. On the other hand, in the tomato 'Santa Clara’ it was observed fewer fruit number, total fresh mass weight and mass average of fruit per plant grown in the soil infested with nematodes (Table 5).

Root-knot nematodes are often associated with high reductions in the productivity of tomato in Brazil. For the cherry tomato plants 'Carolina' and 'Laranja', eggplant, gilo pepper and chili, there were no reductions in the production of plants grown in soil infested when compared with the plants grown in uninfested soil. An exception was observed for tomato 'Santa Clara', which showed high susceptibility to the pathogen.

As for productivity, it is observed in Figure 1 that, with the exception of tomato 'Santa Clara', in which it was evident the negative influence of infestation by root-knot nematode, the other species and cultivars there was no difference between plants propagated in nematode-infested or non-infested soil types. This is suggestive of a measure of tolerance.

According to Sikota and Fernandez (2005), the nematodes of the genus Meloidogyne have been one of the main soil pathogens that commonly affect vegetable crops. In general, the major symptoms that appear in the shoot of plants are wilts caused by the reduction of the capacity of water absorption by the roots. In more severe infestation conditions, it was observed that nutritional deficiency was caused mainly by reducing the transport of essential nutrients in plants, regulated by the root system damaged. The galls are important because they directly compromise the physiology of the plant, causing, in general, reduction in crop production and reduction in quality of the final product (Abad et al., 2009).

Table 4. Productive characters of species and cultivars of Solanaceae grown in soil infested or not with root-knot nematode.

Species/cultivars	Not infested soil	Infested soil	C.V. (%)
Number of total fruits			
Cherry tomato 'Laranja'	0.80^{ns}	0.79^{ns}	19.59
Cherry tomato 'Carolina'	1.08^{ns}	1.09^{ns}	6.17
Hybrid tomato 'T92'	1.36^{ns}	1.28^{ns}	20.50
Chili 'All Big'	0.48^{ns}	0.50^{ns}	23.40
Pepper 'Cayenne'	0.42^{ns}	0.53^{ns}	27.62
Eggplant 'Comprida Roxa '	1.16^{ns}	1.10^{ns}	19.53
Gilo 'Comprido Grande Rio'	0.95^{b}	1.21^{a}	9.91
Tomato 'Santa Clara'	1.22^{a}	1.10^{b}	7.72
Root mass (g. plant $^{-1}$)			
Cherry tomato 'Laranja'	3.58^{b}	14.63^{a}	24.46
Cherry tomato 'Carolina'	26.08^{b}	37.47^{a}	21.03
Hybrid tomato 'T92'	24.15^{a}	14.02^{b}	33.91
Chili 'All Big'	17.90^{b}	32.77^{a}	19.64
Pepper 'Cayenne'	2.58^{b}	7.30^{a}	25.02
Eggplant 'Comprida Roxa '	48.08^{a}	31.02^{b}	24.24
Gilo 'Comprido Grande Rio'	85.10^{ns}	68.92^{ns}	29.04
Tomato 'Santa Clara'	14.28^{b}	38.95^{a}	34.08

*Means followed by the same lowercase letters do not differ at the level of 5% probability by Scott-Knott test´s. ns- not significant.

Table 5. Productive characters of species and cultivars of Solanaceae grown in soil infested or not with root-knot nematode.

Species/Cultivars	Not infested soil	Infested soil	C.V. (%)
Cherry tomato 'Laranja'	61^{ns}	51^{ns}	25.21
Cherry tomato 'Carolina'	38^{ns}	36^{ns}	17.26
Hybrid tomato 'T92'	25^{ns}	27^{ns}	10.19
Chili 'All Big'	21^{ns}	26^{ns}	18.45
Pepper 'Cayenne'	28^{ns}	27^{ns}	16.72
Eggplant 'Comprida Roxa '	24.00^{ns}	25.50^{ns}	9.35
Gilo 'Comprido Grande Rio'	7ns	5ns	36.90
Tomato 'Santa Clara'	24a	8b	16.58
Pasta fresh total fruits (kg.plant^{-1})			
Cherry tomato 'Laranja'	0.65^{ns}	0.58^{ns}	28.87
Cherry tomato 'Carolina'	0.50^{ns}	0.42^{ns}	25.96
Hybrid tomato 'T92'	2.14^{ns}	1.80^{n}s	20.12
Chili 'All Big'	0.86^{ns}	0.93^{ns}	13.94
Pepper 'Cayenne'	0.09^{ns}	0.10^{ns}	30.61
Eggplant 'Comprida Roxa '	2.92^{ns}	3.28^{ns}	30.42
Gilo 'Comprido Grande Rio'	0.23^{ns}	0.16^{ns}	32.53
Tomato 'Santa Clara'	2.75a	0.64b	26.64

*Means followed by the same lowercase letters do not differ at the level of 5% probability by Scott-Knott test´s. ns- not significant.

For the other species and cultivars studied, both production and productivity parameters showed a good production similarly in both soils infested and uninfested. This suggests that there is a tolerance of those to

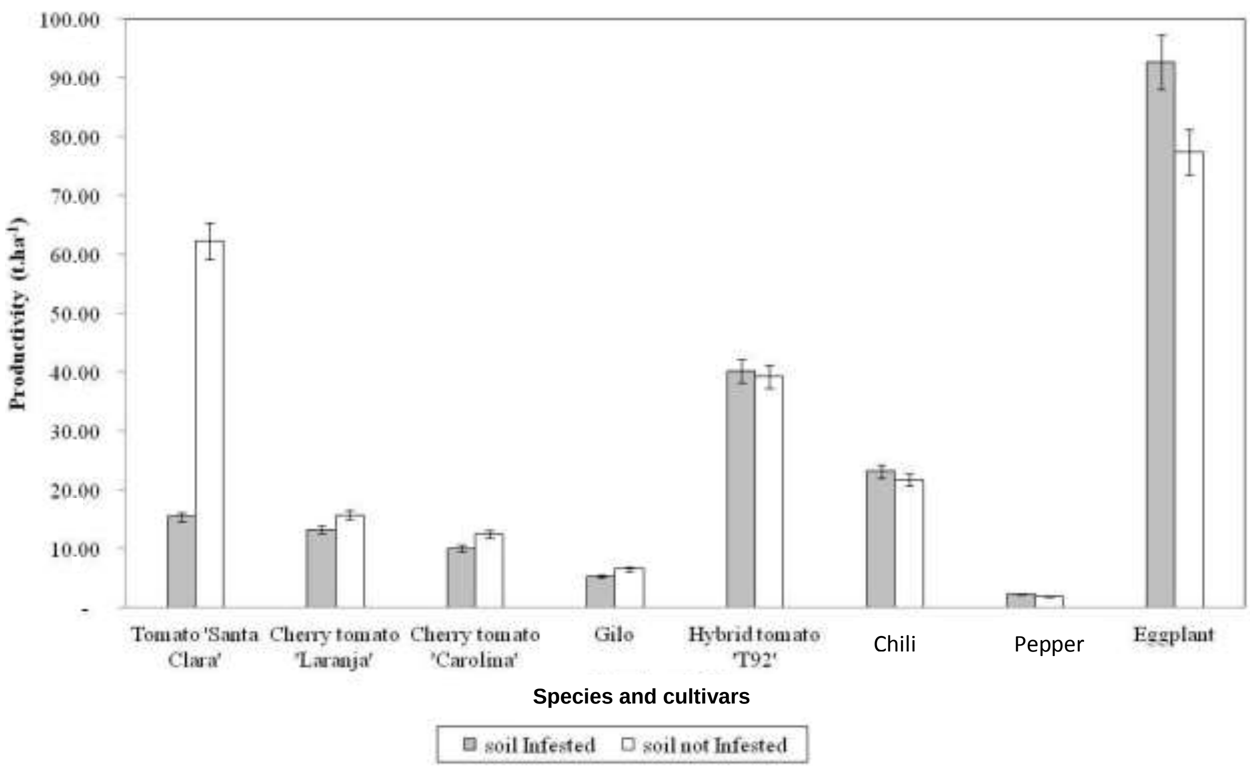

Figure 1. Productivity of species and cultivars of Solanaceae grown in nematode infested or non-infested soil.

infestation by nematode; this is consistent with the non-observation of difference between those physiological factors studied between treatments.

Conclusion

The Pepper 'Cayenne', the Chili 'All Big' and hybrid tomato 'T92' are immune to *M. incognita*. Therefore, they can be used in subsequent studies of resistant – control cultivars using other plant varieties.

The cherry tomato "Laranja" and "Carolina", eggplant 'Comprida Roxa' and Gilo 'Comprido Grande Rio' are tolerant to *M. incognita*, since they were able to produce despite the high infestation. Physiological analysis of the plants must be performed during the production stage, preferably after the start of the fruit harvest, to further evaluate the relationship between the susceptibility to the pathogen and production of the plant.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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