Reaction of soybean cultivars to *Meloidogyne javanica* and *Meloidogyne incognita*

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ABSTRACT

Nematodes of the genus *Meloidogyne* are associated with soybean cultivation, unknown the reaction of cultivars. The objective of this work was to determine the reaction of soybean cultivars to *M. javanica* and *M. incognita*. Twenty-seven soybean cultivars were sown in plastic pots, in a completely randomized experimental design with eight replicates. Ten days after emergence of the seedlings, in a separate experiment, *M. javanica* and *M. incognita* were inoculated. For *M. javanica*, cultivars classified as susceptible were BMX Lança IPRO, AMS Tibagi RR, BMX Vanguarda IPRO, NS 6700 IPRO, BMX Ativa RR, FPS Solimões RR, TEC 6702 IPRO, NS 5909 RR, NS 5445 IPRO, 54i52 RSF IPRO, M 6410 IPRO and NS 5959 IPRO and BMX Elite IPRO, NA 6211 RR, BMX Valente RR, FPS Júpter RR, FPS Iguaçú RR, BMX Tornado RR, GMX Cancheiro RR, TMG 7161 IPRO, TMG 7062 IPRO, FPS Solar RR, M 5730 IPRO, DM5958 RSF IPRO, NS 6006 IPRO, SYN 1163 RR and M 5947 IPRO were resistant. Regarding *M. incognita*, the resistant cultivars were FPS Iguaçú RR, FPS Solar IPRO, SYN 1163 RR and TMG 7062 IPRO. The cutlivars FPS Júpter RR, SYN 1163 RR e TMG 7062 IPRO were resistant to both nematodes.

Keywords: reproduction factor; resistance; root-knot nematode; susceptibility.

INTRODUCTION

Phytonematodes are soil pathogens. Their control is difficult to be achieved and requires high costs for Brazilian farming production. The nematodes of the genus *Meloidogyne* are among the phytonematodes most frequently associated with soybean cultivation with a wide geographic distribution. They are responsible for raising the losses, mainly caused by the expansion of soybean cultivation to new agricultural frontiers, intensification of monoculture and the adoption of inadequate management practices of these pathogens (Juhász *et al*., 2013). For this genus, more than 80 species are described, and the main ones for the soybean crop are *Meloidogyne javanica* (Treub, 1885) and *Meloidogyne incognita* (Kofoid & White, 1919) Chitwood, 1949 (Miranda *et al*., 2011). The most common species is *M. javanica*, which has a widespread occurrence and causes losses of 10% to 40% in sandy or medium-sandy soils. This nematode predominates in areas previously cultivated with coffee or cotton and is associated with succession of soybean-cotton and soybean-corn crops (Miranda *et al*., 2011).

The typical symptom of an attack by a *Meloidogyne* nematode is hyperplasia, that is, thickening of the cells of the root cortex named gall. As a reflex symptom, the leaves of the attacked plants may show yellowing due to nitrogen deficiency and also chlorotic spots or necrosis between the veins, which characterizes "carijó" leaf (Deuner *et al*., 2012).

For a successful controlling of phytonematodes, several alternatives can be used, but most of them are limited by factors such as: habitat type (soil and root interior), morphological characteristics (presence of resistant cuticle), and polyphagous habit of feeding on several plant

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species (Soares *et al*., 2016). In the integrated management, associated strategies such as cultural control (crop rotation), genetic control (resistant cultivars), chemical control (nematicides) and biological control (fungi and bacteria) should be used (Almeida *et al*., 2005). Therefore, the plant genetic improvement has been developing soybean cultivars with high productivity, wide adaptation and good resistance/tolerance to pathogens. Genetic resistance is one of the best ways to control nematodes that is why it has easy assimilation by farmers, does not increase production costs and does not environmental cost of using pesticides (Teixeira, 2013).

In spite of this, many breeders do not have the information of the reaction of the cultivar to the nematodes, although this information is of fundamental importance to compose the integrated management of nematodes. There are still several cultivars whose responses (resistant or susceptible) to the nematodes of the genus *Meloidogyne* are still not known.

Therefore, the objective of this work is to determine the reaction of soybean cultivars to *M. javanica* and *M. incognita*.

MATERIAL AND METHODS

Two experiments were carried out, one for each nematode, where 27 soybean cultivars were selected (Table 1), indicated for the southern region of Brazil, for which no information is available or there is partial information on the reaction to *M. javanica* and *M. incognita* by the breeding companies of the cultivars. Crotalaria (*Crotalaria spectabilis* Roth.) and tomato (*Solanum lycopersicum* L.) cv. Santa Cruz Kada were included in the experiment as resistance and susceptibility patterns, respectively.

In order to conduct the experiment, the seeds of the soybean cultivars were sown in plastic containers with a capacity of 5 L using a mixture of sterilized sand and soil (3: 1).

The populations of *M. javanica* and *M. incognita* were originated from the municipalities of Piacatú/SP and Barreiras/BA, respectively. They were purified and multiplied in cv. Santa Cruz Kada tomato plants and kept in a greenhouse. Ten days after the emergence of soybean plants (DAE), manual thinning was performed, maintaining two plants per pot. Each plant was inoculated with a suspension of 5000 eggs and juveniles of second stage (J2) per mL. The inoculum was obtained by washing and grinding the roots of tomato in sodium hypochlorite solution (0.5%), according to the methodology described by Hussey & Barker (1973) and adapted by Bonetti & Ferraz (1981). The egg suspension and J2 were deposited in a hole about 3 cm deep next to each plant in the pot. After inoculation, the plants were maintained for 90 days in a greenhouse and watered daily, maintaining the adequate moisture level in the soil for plant growth. The temperature in this period in the greenhouse ranged from 14 to 32 °C.

After 90 days, the eggs and J2 present in the root system of each plant were extracted according to methodology of Hussey & Barker (1973) adapted by Bonetti & Ferraz (1981). This same procedure was performed for each of the nematode species in the evaluated soybean cultivars. The number of eggs and J2 present in the suspension of each replicate were counted using an optical microscope (magnification 100x) with the aid of a Peters chamber. Subsequently, the Reproduction Factor (RF) was determined by the division of the Final Population (FP) by the Initial Population (PI) (Oostenbrink, 1966). Behavior of each cultivar was classified according to the criterion established by Oostenbrink (1966), and cultivars that obtained RF greater than or equal to 1 were considered susceptible, whereas those that obtained RF less than 1 were considered resistant.

The experiments were conducted in a completely randomized experimental design with eight replicates. The data achieved in the experiment were transformed into square roots of X + 0.5 and submitted to analysis of variance. The averages were grouped by the Scott-Knott test at 5% probability of error through the statistical program Assistat version 7.7 beta (pt) (Silva & Azevedo, 2002).

RESULTS AND DISCUSSION

In the *M. javanica* nematode test (Table 2), the final nematode population (FP) of the soybean cultivars ranged from 250 to 13,044 individuals per plant, and this was observed for cultivars M 5947 IPRO and BMRO IPRO, respectively. The standard of susceptibility (tomato) used had a final population (FP) of 188.856 and resistance (crotalaria) of 40, with a reproductive factor (RF) of 37.8 and 40, respectively.

Based on the final population, it was verified that the highest value of this variable was obtained in the BMX cultivars IPRO, AMS Tibagi RR, BMX Vanguarda IPRO, NS 6700 IPRO, BMX Ativa RR and SPS Solimões RR, with no statistical difference between them, but with difference for the other cultivars, tomato and crotalaria. The lowest final population value was observed for cultivar M 5947 IPRO, which did not differ statistically from crotalaria, but differed from other cultivars and from tomato

In relation to the reproduction factor (RF), and by taking into account the cultivar classification criterion proposed by Oostenbrink (1966), the reaction of soybean cultivars to *M. javanica* can be separated into two groups (Table 2). The first one is made up by susceptible cultivars that had RF between 1.0 and 2.6, being these BMX Lance IPRO, AMS Tibagi RR, BMX Vanguard IPRO, NS 6700 IPRO, BMX Active RR, FPS Solimões RR, TEC 6702 IPRO, NS 5909 RR, NS 5445 IPRO, 54i52 IPRO RSF, M 6410 IPRO and NS 5959 IPRO. In the second group, the resistant cultivars with RF between 0 and 0.8, which were BMX Elite IPRO, NA 6211 RR, BMX Valente RR, FPS Jupter RR, FPS Iguaçú RR, BMX Tornado RR, GMX Cancheiro RR, TMG 7161 IPRO , TMG 7062 IPRO, FPS Solar RR, M 5730 IPRO, DM5958 IPRO RSF, NS 6006 IPRO, SYN 1163 RR and M 5947 IPRO. Therefore, it was observed that 44.4% of the cultivars were considered susceptible, while 55.6% were resistant to *M. javanica*.

Regarding *M. incognita* (Table 3), the final population in the soybean cultivars varied from 2,995 to 34,600 individuals per plant. These results were observed for cultivars TMG 7062 IPRO and NS 6211 RR, respectively. The susceptibility pattern (tomato) had a PF of 137.036 and the resistance (crotalaria) of 50 with RF of 27.4 and 0.0, respectively. The largest value of this variable was verified for the cultivar NS 6211 RR, which differed statistically from the other cultivars and the resistance and susceptibility patterns. The lowest value was for the cultivars FPS Iguaçú RR, FPS Solar IPRO, SYN 1163 RR and TMG 7062 IPRO.

Two groups were separated according to the cultivar classification criterion proposed by Oostenbrink (1966). The first group refers to the group of susceptible cultivars (RF of 1.0 to 6.9), and the second group was composed of the resistant ones (RF between 0.6 and 0.8). In the second group were the cultivars FPS Iguaçú RR, FPS Solar IPRO, SYN 1163 RR and TMG 7062 IPRO, and in the first group, the other cultivars. Thus, out of the 27 soybean cultivars evaluated, 85.2% were susceptible and 14.8% were resistant to M. incógnita.

Cultivars FPS Jupiter RR, SYN 1163 RR and TMG 7062 IPRO were classified as resistant to both nematodes.

According to Faria *et al*. (2003), one of the mechanisms of resistance is the accumulation of phytoalexin in resistant hosts, which coincides with the hypersensitivity reaction, thus functioning as nematostatic phytoalexins, drastically affecting nematode function and preventing its development. In studies by Mattos (2013), hostability, aggressiveness and virulence are factors that interfere with the interspecific variation of *Meloidogyne* species in the plant-nematoid interaction. According to Silva (2001), the suppression of nematode development and reproduction varies according to the resistance and susceptibility of the plants, and plants that are considered highly resistant allow very low reproduction rates, while

Table 1: Description of commercial soybean cultivars with their respective characteristics

Cultivar	Breeder	Growth habit	Maturation Groupo
54i52 RSF IPRO	GDM Genética do Brasil	Undetermined	5.4
AMS Tibagi RR	Bayer S.A.	Semi-Determined	5.0
BMX Ativa RR	Brasmax	Determined	5.6
BMX Elite IPRO	Brasmax	Undetermined	5.5
BMX Lança IPRO	Brasmax	Undetermined	5.8
BMX Tornado RR	Brasmax	Undetermined	6.2
BMX Valente RR	Brasmax	Undetermined	6.7
BMX Vanguarda IPRO	Brasmax	Undetermined	6.0
DM RSF 5958 IPRO	Don Mario	Undetermined	5.8
FPS Iguaçu RR	Fundação Pró-sementes	Undetermined	5.0
FPS Júpiter RR	Fundação Pró-sementes	Undetermined	5.9
FPS Solar IPRO	Fundação Pró-sementes	Undetermined	6.3
FPS Solimões RR	Fundação Pró-sementes	Undetermined	5.7
GMX Cancheiro RR	Gmax Genética	Undetermined	6.2
M 5730 IPRO	Monsoy	Undetermined	5.7
M 5947 IPRO	Monsoy	Undetermined	5.9
M 6410 IPRO	Monsoy	Undetermined	6.4
NS 5445 IPRO	Nidera	Undetermined	5.4
NA 5909 RG	Nidera	Undetermined	6.2
NS 5959 RG	Nidera	Undetermined	6.9
NS 6006 IPRO	Nidera	Undetermined	5.7
NS 6211 RR	Nidera	Determined	6.2
NS 6700 IPRO	Nidera	Undetermined	7.1
SYN 1163 RR	Syngenta	Undetermined	6.3
TEC 6702 IPRO	CCGL TEC	Undetermined	6.7
TMG 7062 IPRO	TMG[1]	Semi-determined	6.2
TMG 7161 RR	TMG[1]	Undetermined	5.9

[1]Tropical Melhoramento e Genética; Source: Registro Nacional de Cultivares (MAPA).

the susceptible ones allow nematodes to reproduce abundantly.

In a study developed by Moritz *et al*. (2008), it was observed on the eighth day after inoculation that most of the nematodes that had penetrated the roots of the susceptible and resistant soybean cultivars were still at the J2 developmental stage, remaining parallel to the central cylinder of the roots. This could indicate that resistance mechanisms of the resistant cultivar were not yet slowing the overall development of the nematode. Similarly, Moura *et al*. (1993) observed that in soybean cultivars resistant to *M. incognita*, the root cells became disorganized and necrotic, and the formation of the feeding sites did not occur, preventing the development of the nematode to the J3 stage 10 days after inoculation.

In a study by Dalla Favera (2014), it was verified that out of the 45 soybean cultivars evaluated, in order to determine the reaction to *M. javanica*, all were susceptible. Furthermore, the highest density of phytonematodes per gram of roots was obtained in cultivar AMS Tibagi RR., data similar to those of the present study, where this cultivar proved to be one of the most susceptible for both *M. javanica* and *M. incognita*. Among the six soybean cultivars evaluated by Kirsch (2016), all presented RF greater than 1, being classified as susceptible to *M. javanica*; however, the cultivar BMX Turbo RR showed to be resistant to some populations of *M. javanica*, with RF less than 1. In studies by Santos & Soares (2009), although many soybean cultivars were considered susceptible, they showed values of FR close to 1, which is less susceptible to *M. javanica* and *M. arenaria* (Neal, 1889) Chitwood, 1949. Therefore, it is suggested that in the absence of cultivars resistant to root-knot nematodes, cultivars with lower FR should be used because of the lower susceptibility presented by them. According to Alves *et al*. (2011), cultivars with high RF or greater than one, are susceptible and should be avoided in areas with nematode presence, especially

Table 2: Nematode final population (egg and J2/plant), reproduction factor (RF) and reaction (R) of twenty-seven soybean cultivars to *Meloidogyne javanica* at 90 days after inoculation

Cultivar	Final population (FP)	Reproduction factor (FR)	Classification[4]
Tomate[1]	188,856 a[2,3]	37.8	Susceptible
BMX Lança IPRO	13,044 b	2.6	Susceptible
AMS Tibagi RR	12,630 b	2.5	Susceptible
BMX Vanguarda IPRO	11,635 b	2.3	Susceptible
NS 6700 IPRO	11,262 b	2.3	Susceptible
BMX Ativa RR	11,117 b	2.2	Susceptible
FPS Solimões RR	9,391 b	1.9	Susceptible
TEC 6702 IPRO	8,090 c	1.6	Susceptible
NS 5909 RR	8,086 c	1.6	Susceptible
NS 5445 IPRO	7,155 c	1.4	Susceptible
54i52 RSF IPRO	6,692 c	1.3	Susceptible
M 6410 IPRO	6,695 c	1.3	Susceptible
NS 5959 IPRO	4,822 d	1.0	Susceptible
BMX Elite IPRO	3,865 e	0.8	Resistant
NA 6211 RR	3,680 e	0.7	Resistant
BMX Valente RR	3,442 e	0.7	Resistant
FPS Jupiter RR	3,405 e	0.7	Resistant
FPS Iguaçú RR	2,115 f	0.4	Resistant
BMX Tornado RR	2,1025 f	0.4	Resistant
GMX Cancheiro RR	2,002 f	0.4	Resistant
TMG 7161 IPRO	1,824 f	0.4	Resistant
TMG 7062 IPRO	1,800 f	0.4	Resistant
FPS Solar RR	1,760 f	0.4	Resistant
M 5730 IPRO	1,350 f	0.3	Resistant
DM5958 RSF IPRO	887 g	0.2	Resistant
NS 6006 IPRO	810 g	0.2	Resistant
SYN 1163 RR	797g	0.2	Resistant
M 5947 IPRO	250 h	0.0	Resistant
Crotalária[5]	40 h	0.0	Resistant
C.V. (%)	14.6	-	-

Note: [1]Suceptibility pattern. [2]Data transformed into square root of $x+0.5$. [3]Means followed by the same letter in the columns belong to the same group (Scott; Knott, 5% of probability). [4]Classification proposed by Oostenbrink (1966); [5]Resistence pattern.

M. javanica and *M. incognita* species. However, caution must be exercised in the behavior of susceptibility of cultivars, since in addition to the genetic characteristics of the material to be recognized by the nematode and used as a food source, it may be linked to the environmental conditions in which the crop is found (Li & Chen, 2005).

In a work associating control strategies, Araujo *et al*. (2012) concluded that only the use of a resistant soybean cultivar to *M. javanica* and *M. incognita* did not reduce the incidence of nematodes in the roots neither it increased the dry mass of the aerial part of the plants. On the other hand, the association of resistant cultivars with chemical or biological control provided a reduction in the incidence of nematodes in the roots, and enhanced soybean growth. Similarly, Kamunya *et al*. (2008) reported that the use of resistant genotypes is undoubtedly the best option, since it is of low cost and agroecologically correct to control nematodes, but nevertheless, the use of other associated techniques can provide better control results.

Information on Brazilian cultivars resistant to the species *M. javanica* and *M. incognita* can be found in the literature, but often in a partial and contradictory way, depending on the criteria used by the researchers (Mendes & Rodriguez, 2000). Considering the results presented by Kirsch (2016), the author verified that resistance sources are available among the soybean cultivars, for the root-knot nematode species evaluated in her work. However, she emphasizes that there are few evaluated sources of resistance and that the levels are not high. Therefore, the development of a work related to this subject requires accurate and continuous information, since soybean cultivars are made available in the market every year and information on the reaction of the cultivar is one of the main management strategies of the root-knot nematodes.

Table 3: Nematode final population (egg and J2/plant), reproduction factor (RF) and reaction (R) of twenty-seven soybean cultivars to Meloidogyne incognita at 90 days after inoculation

Cultivar	Final Population (FP)	Reproduction Factor (RF)	Classification[4]
Tomato[1]	137.036 a[2,3]	27.4	Susceptible
NS 6211 RR	34.600 b	6.9	Susceptible
BMX Valente RR	26.616 c	5.3	Susceptible
AMS Tibagi RR	25.517 c	5.1	Susceptible
BMX Tornado RR	24.825 c	4.9	Susceptible
NS 6006 IPRO	24.592 c	4.9	Susceptible
M 5947 IPRO	22.975 c	4.6	Susceptible
FPS Solimões RR	20.908 d	4.2	Susceptible
M 5730 IPRO	19.890 d	3.9	Susceptible
BMX Vanguarda IPRO	15.575 e	3.1	Susceptible
54i52 RSF IPRO	15.166 e	3.0	Susceptible
NS 6410 IPRO	14.915 e	2.9	Susceptible
BMX Elite IPRO	12.300 f	2.5	Susceptible
TEC 6702 IPRO	12.136 f	2.4	Susceptible
BMX Ativa RR	11.160 f	2.2	Susceptible
FPS Júpter RR	10.280 f	2.0	Susceptible
NS 5445 IPRO	9.135 f	1.8	Susceptible
TMG 7161 IPRO	9.085 f	1.8	Susceptible
NS 5909 RR	8.961 f	1.8	Susceptible
GMX Cancheiro RR	7.065 g	1.5	Susceptible
BMX Lança IPRO	6.750 g	1.3	Susceptible
DM 5959 IPRO	6.367 g	1.3	Susceptible
DM5958 RSF IPRO	6.066 g	1.2	Susceptible
NS 6700 IPRO	5.250 g	1.0	Susceptible
FPS Iguaçú RR	4.047 h	0.8	Resistant
FPS Solar IPRO	3.653 h	0.7	Resistant
SYN 1163 RR	3.025 h	0.6	Resistant
TMG 7062 IPRO	2.995 h	0.6	Resistant
Crotalária[5]	50 I	0.0	Resistant
C.V.	9.2	-	-

Note: [1]Susceptibility pattern; [2]Data transformed into square root of x+0.5. [3]Means followed by the same letters in the column belong to the same group (Scott Knott, 5% of probability). [4]Classification proposed by Oostenbrink (1966).

CONCLUSIONS

The cultivars BMX Lança IPRO, AMS Tibagi RR, BMX Vanguarda IPRO, NS 6700 IPRO and BMX Ativa RR are susceptible to *M. javanica* and cultivars M 5947 IPRO, SYN 1163 RR, NS 6006 IPRO, DM5958 RSF IPRO, M 5730 IPRO, FPS Solar RR, TMG 7062 IPRO, TMG 7161 IPRO, GMX Cancheiro RR, BMX Tornado RR, FPS Iguaçú RR, FPS Júpter RR, BMX Valente RR, NA 6211 RR e BMX Elite IPRO are resistant.

Regarding *M. incognita*, the cultivar NS 6211 RR is susceptible and the cultivars TMG 7062 IPRO, SYN 1163 RR, FPS Solar RR and FPS Iguaçu RR are resistant.

Cultivars TMG 7062 IPRO, SYN 1163 RR, FPS Solar RR and FPS Iguaçú RR are resistant to *M. javanica* and *M. incognita*.

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