



Host status of selected summer forage grasses to *Meloidogyne javanica* and *M. incognita*

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Abstract

Forage grasses have been included in systems (no-tillage) with crops susceptible to important plant parasitic nematodes (PPNs), but the host status of commercial genotypes is still poorly known. Therefore, the objective of our study was to evaluate the host status of summer forage species to *Meloidogyne javanica* (*Mj*) and *M. incognita* (*Mi*), under greenhouse conditions. For each interaction, the trials were repeated twice. For this, seedlings were inoculated with an initial population (*Ip*) of 1,000 specimens (eggs plus second-stage juveniles). After 60–71 days of inoculation, the specimens were extracted from the roots (*Fp*) to obtain estimates of the reproductive factor ($RF = Fp/Ip$) and nematodes per gram root ($Nema\ g^{-1}$). The species/cultivars tested were Millet ‘ANM 38’, *Brachiaria ruziziensis* ‘BRS Integra’, *B. ruziziensis* x *B. brizantha* ‘BRS RB331 Ipyporã’, *B. brizantha* ‘BRS Piatã’, *Panicum maximum* ‘BRS Tamani’, *P. maximum* ‘BRS Quênia’ and *P. maximum* ‘BRS Zuri’. All species/cultivars tested were rated as resistant to *M. javanica* ($0 \leq RF \leq 0.19$; $0 \leq Nema\ g^{-1} \leq 141.44$) and *M. incognita* ($0.01 \leq RF \leq 0.07$; $2.43 \leq Nema\ g^{-1} \leq 66.74$). Our study opens the possibility of using these forage species/cultivars for rotation/succession schedules in areas with infestation by these PPNs.

Keywords Cultural control · Crop rotation and succession · Root-knot nematodes · Southern Brazil

Brazil has approximately 160 million hectares of pastures, including native and cultivated species (Antonio 2015). In recent years, Brazil showed an increase in pasture cultivation, encompassing about 70% of the cropped area in Brazil (Omote et al. 2021). The use of forages in Brazil has been increasingly advancing due to the benefits related to the production system, such as the no-tillage system (Inomoto et al. 2007). Therefore, the use of forages has become increasingly important in the intensification of agricultural production systems, whether in forage production (crop-livestock system) or the production of dry matter for soil coverage and improving the structural quality of the soil (Silva et al. 2015; Assis et al. 2018). Furthermore, some plant species have historically played an important role (poor host or non-host)

in the integrated management of the major plant parasitic nematodes (PPNs), influencing the dynamics of PPN populations (Asmus and Cruz 2020; Gabriel et al. 2018).

Meloidogyne species are undoubtedly the main group of PPNs worldwide, known as root-knot nematodes (RKNs) (Trudgill and Blok 2001). Of the ninety species of RKNs described, *Meloidogyne incognita* (*Mi*) (Kofoid & White) Chitwood and *M. javanica* (*Mj*) (Treub) Chitwood stand out by their high geographical distribution and/or damage intensity (Elling 2013). For these reasons, it is necessary to develop new tools for reducing their populations in infested areas with *Mj* and *Mi*. Within this framework, it is extremely important to evaluate the host status of poaceous forage cultivars to the major *Meloidogyne* species, so that they can be used in areas cultivated with cropping systems affected by these PPNs, such as those with soybean. It should be noted that *Mj* and *Mi* have historically been associated with significant losses in soybeans (Gorny et al. 2021), with estimated reductions of over 80% (Asmus and Ferraz 2001). Therefore, the objective of our study was to determine the host status of selected forager grasses to *Mj* and *Mi* under greenhouse conditions.

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The *Mj* and *Mi* nematode isolates (one egg-masses) were previously identified from esterase (Est) phenotypes (Carneiro and Almeida 2001) and maintained on tomato [*Solanum lycopersicum* (L.)] ‘Santa Clara’. The isolate of *Mj* (Est J3) came from Tupanciretã (RS) while *Mi* (Est I1) of Urusanga (SC). All trials were performed twice under greenhouse conditions at the Universidade Federal de Pelotas, Capão do Leão, Rio Grande do Sul state. The trials were conducted between March 05, 2022, and May 14, 2022 [trial 1 (*Mj*)], March 09, 2022, and May 7, 2022 [trial 2 (*Mi*)], and their repetitions were conducted between February 24, 2024, and April 26, 2024 [trial 1 (*Mj*) and trial 2 (*Mi*)].

Initially, seeds of *Panicum maximum* (Jacq.) ‘BRS Quênia’, *P. maximum* ‘BRS Tamani’, *P. maximum* ‘BRS Zuri’, millet [*Pennisetum glaucum* (L.) R] ‘ANM 38’, *B. ruziziensis* x *B. brizantha* ‘BRS RB331 Ipyporã’, *B. brizantha* (Stapf) ‘BRS Piatã’ and *B. ruziziensis* (Germ & Evrard) ‘BRS Integra’ were germinated in boxes with filter paper soaked in distilled water. After five days, three seedlings were transplanted into each pot (1000 ml). Subsequently, we leave only one plant per pot. Tomato ‘Santa Cruz Kada’ was included as control of susceptibility. The composition of the substrate used consisted of a mixture (1:1) of commercial substrate and previously autoclaved soil (120°C for 2 h). Unfortunately, millet was not included in the second evaluation because of unavailability of seeds.

The initial populations (*Ip*) were of 1000 specimens [eggs plus juveniles of second stage (J2s)] and introduced into two (2 cm-deep) holes around seedlings. The final population (*Fp*) was obtained at 71 (trial 1—2022), 60 (trial 2—2022) and 62 (trials 1 and 2—2024) *dai*, according to Coolen and D’Herde (1972) using sodium hypochlorite 0.5%. Nematode resistance was measured by the reproductive factor

($RF = Fp/Ip$) and number of nematodes per gram of root (*Nema g*⁻¹) and defined according to statistical analysis. We consider plants/cultivars to be resistant if the RF is less than 1 and to be susceptible if the RF is greater than 1 (Oostenbrink 1966). All the trials were carried out in a completely randomized design and five replicates. Each experimental unit was composed of one plant per pot.

Data were analyzed by one-way analysis of variance (ANOVA) and means of the treatments compared by Tukey’s HSD test ($P \leq 0.01$) using software R 4.2.1 (R Development Core Team 2022). The data set was transformed into $\ln(x + 1)$ before ANOVA (Noe 1985).

The high RF values observed for tomatoes prove the viability of the inoculum used and the adequacy of the experimental conditions adopted for both PPNs (Tables 1 and 2). All tested species/cultivars were resistant to *Mj* ($RF < 1.0$) (Table 1) ($P < 0.01$). Similarly, the same trend was found for trials with *Mi*, in which the RF values were lower than 1, being rated as resistant (poor host) (Table 2) ($P < 0.01$). The results of *Nema g*⁻¹ strongly corroborate the results obtained for RF (similar trend), whether for *Mj* ($0 \leq Nema g^{-1} \leq 141.44$) or *Mi* ($2.43 \leq Nema g^{-1} \leq 66.74$). The results presented here highlight the strong response of the tested species/cultivars, strengthening the reliability of the data obtained and signaling the promising use. Galls were not observed for any grasses, regardless of the year.

In a study carried out by Carneiro et al. (2006), the obtained RF values for all *Brachiaria* species/cultivars were less than 1.0, which means a considerable reduction in the population of these PPNs. For instance, *B. brizantha* ‘MG5’, *B. brizantha* ‘Marandu’, *B. decumbens* x *B. brizantha* ‘MG4’ showed resistance to *Mj* and *Mi*. In this same study, *P. maximum* ‘Tanzânia’ and *P. maximum* ‘Mombaça’

Table 1 Reproductive factor (RF), number of nematodes per gram of roots (*Nema g*⁻¹) and host status for forage species/cultivars inoculated (1000 specimens) with *Meloidogyne javanica* under greenhouse conditions

Treatments	Trial 1 (2022)		Trial 2 (2024)		Host status ³
	RF	<i>Nema g</i> ⁻¹	RF	<i>Nema g</i> ⁻¹	
Tomato ¹	13.51 ± 6.41 a ²	3,637.11 ± 1,821.75 c	44.13 ± 3.35 a	20,328.1 ± 2,722.1 a	S
<i>Panicum maximum</i> ‘BRS Tamani’	0 ± 0 b	0 ± 0 a	0.06 ± 0.02 b	18.85 ± 12.24 b	R
Millet ‘ANM 38’	0.19 ± 0.21 b	105.53 ± 115.24 b	-	-	R
<i>B. ruziziensis</i> x <i>B. brizantha</i> ‘BRS RB331 Ipyporã’	0 ± 0 b	0 ± 0 a	0.04 ± 0.02 b	20.06 ± 6.85 b	R
<i>Brachiaria brizantha</i> ‘BRS Piatã’	0 ± 0 b	0 ± 0 a	0.03 ± 0.02 b	9.79 ± 4.23 b	R
<i>Panicum maximum</i> ‘BRS Quênia’	0 ± 0 b	0 ± 0 a	0.07 ± 0.02 b	14.7 ± 5.39 b	R
<i>Brachiaria ruziziensis</i> ‘BRS Integra’	0.11 ± 0.02 b	141.44 ± 101.26 b	0.06 ± 0.03 b	42.18 ± 33.01 b	R
<i>Panicum maximum</i> ‘BRS Zuri’	0 ± 0 b	0 ± 0 a	0.03 ± 0.02 b	66.74 ± 55.96 b	R

¹ Control of susceptibility

² Means (± standard deviation) followed by the same letter in each column did not differ significantly ($P = 0.01$) by Tukey’s HSD test

³ S = susceptible ($RF > 1.0$); R = resistant ($RF < 1.0$); — = not tested

n = 5 replicates for each trial

Table 2 Reproductive factor (RF), number of nematodes per gram of roots (Nema g⁻¹) and host status of forage species/cultivars inoculated (1000 specimens) with *Meloidogyne incognita* under greenhouse conditions

Treatments	Trial 1 (2022)		Trial 2 (2024)		Host status
	RF	Nema g ⁻¹	RF	Nema g ⁻¹	
Tomato ¹	26.23 ± 7.82 a	5,232.06 ± 2,363.81 a	18.80 ± 6.13 a	5,996.73 ± 2,622.52 a	S
<i>Panicum maximum</i> ‘BRS Tamani’	0.12 ± 0.20 b	29.99 ± 44.26 b	0.01 ± 0.02 b	3.52 ± 3.76 bc	R
Millet ‘ANM 38’	0.09 ± 0.13 b	45.56 ± 60.02 b	-	-	R
<i>B. ruziziensis</i> x <i>B. brizantha</i> ‘BRS RB331 Ipyporã’	0 ± 0 b	0 ± 0 b	0.06 ± 0.03 b	31.26 ± 23.04 c	R
<i>Brachiaria brizantha</i> ‘BRS Piatã’	0.06 ± 0.06 b	37.31 ± 50.54 b	0.02 ± 0.02 b	15.38 ± 13.73 bc	R
<i>Panicum maximum</i> ‘BRS Quênia’	0.01 ± 0.01 b	1.10 ± 1.58 b	0.05 ± 0.03 b	18.16 ± 15.80 bc	R
<i>Brachiaria ruziziensis</i> ‘BRS Integra’	0 ± 0 b	0 ± 0 b	0.01 ± 0.02 b	2.43 ± 3.75 b	R
<i>Panicum maximum</i> ‘BRS Zuri’	0 ± 0 b	0 ± 0 b	0.01 ± 0.01 b	6.04 ± 6.03 bc	R

¹ Control of susceptibility² Means (± standard deviation) followed by the same letter in each column did not differ significantly ($P=0.01$) by Tukey's HSD test³ S = susceptible (RF > 1.0); R = resistant (RF < 1.0); — = not tested $n=5$ replicates for each trial

showed RF values less than 1.0. Similar results were observed by Dias-Arieira et al. (2003) for *P. maximum*, *B. brizantha* ‘MG4’ and *B. decumbens*. Still, these results are aligned with the work of Brito and Ferraz (1987), in which *B. decumbens* and *P. maximum* ‘Guiné’ reduced the population of *Mj*. Brito and Ferraz (1987) verified also that this antagonism was due to the lower rate of hatching and development of J2s.

In relation to millet, according to research conducted by Nascimento et al. (2020), the results revealed that the exclusive use of millet ‘ADR 300’ provided a significant reduction of 61% and 72% in the population levels of both PPNs. On the other hand, Chidichima et al. (2021) evaluated the host status of millet cultivars (‘ADR300’, ‘ADR500’ and ‘BRS 1501’) to four *Mj* isolates and the authors verified that all studied cultivars were susceptible to *Mj*. This variability was verified also as Inomoto et al. (2008), in which millet ‘ADR300’ was rated as susceptible while millet ‘BN2’ was resistant to *Mj*. Notwithstanding, it is necessary to mention discrepant results, for which variable reactions were identified depending on the race of *Mi* inoculated (Carneiro et al. 2007).

Taken together, our findings will help the appropriate selection of poaceous forage species/cultivars to be used in crop rotation and succession, contributing to the efficient management of PPNs the southern Brazil. Indeed, according to surveys conducted by Kirsch et al. (2016) and Marquez et al. (2021), *M. javanica* was the major *Meloidogyne* species in soybean-producing regions in the state of Rio Grande do Sul. In relation to *Mi*, it is known that it is widely distributed in other states in Brazil (Inomoto et al. 2008). In this sense, all the tested species/cultivars may be introduced in these areas, resulting in considerable populational reduction.

Furthermore, resistance to these PPNs should be considered in future breeding programs.

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Data availability The dataset was obtained from trials carried out by the authors and is available from the corresponding author.

Declarations

Conflicts of interests The authors declare no conflict of interest.

Research involving humans and/or animals Not applicable.

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