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Host Status of Brazilian Native Tree Species to Root-Knot Nematodes

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ABSTRACT

Root-knot nematodes (RKN, *Meloidogyne* spp.) are among the most important plant pathogenic organisms, causing significant damage, with a wide geographical distribution and being difficult to control. The ability of these nematodes to parasitize native trees from Brazilian biomes is little understood. This study evaluated the host status of 24 native tree species to *M. incognita* and *M. javanica* under greenhouse conditions. Overall, 62.5% of the species evaluated (15 out of 24) were classified as susceptible (reproduction factor, RF > 1.0) to *M. javanica*, whereas 70.8% (17 out of 24) were susceptible to *M. incognita*. The highest RF values for both nematodes were recorded in *Enterolobium contortisiliquum*, *Handroanthus albus* and *Senna macranthera*, while *Cedrela fissilis* showed high susceptibility only to *M. incognita*. Six species were resistant to *M. javanica* and five to *M. incognita*, with four species (*Campomanesia guazumifolia*, *Erythrina falcata*, *Inga edulis* and *Parapiptadenia rigida*) exhibiting resistance to both nematodes. In addition, four species were immune to *M. javanica* and two to *M. incognita*, with *Apuleia leiocarpa* and *Myrcianthes pungens* being the only species immune to both RKN species. Many of the most susceptible trees are widely distributed in the Cerrado, Brazil's leading grain- and fibre-producing region, indicating potential refuges for RKN near agricultural landscapes. This study provides the first comprehensive evaluation of native Brazilian trees as RKN hosts, filling a major knowledge gap and offering novel insights for sustainable nematode management, forest restoration and biotechnological exploration.

1 | Introduction

Studies on the diversity of plant-parasitic nematodes, in agricultural areas and areas covered by primary vegetation have aroused growing interest, due to the progressive increase in economic losses caused on agricultural activity by these pathogens (AbdelRazek and Balah 2023). Regarding the plant-parasitic nematodes community in natural ecosystems, the studies are justified in biological terms by the great representativeness of the phylum Nematoda. In economic terms, the justification lies in its possible effects on vegetation and, mainly, on the need to understand the ecology of plant-parasitic nematodes in their

original environment, since species of agricultural importance are nothing more than organisms originating from natural ecosystems that are adapted to cultivated areas (Costa et al. 2011; Ramos et al. 2023).

Plant-parasitic nematodes (PPN) belonging to the genus *Meloidogyne*, commonly known as root-knot nematodes (RKN), are among the major pathogenic agents that cause economic damage to plants (Bellé et al. 2020). Some species of this genus are distributed worldwide and are able to parasitize a wide range of plants from different botanical families (Ralmi et al. 2016). They represent the most important group of PPN worldwide,

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with more than 105 species described (Ghaderi and Karssen 2020) and a wide range of host plants, posing a serious threat to global agricultural production.

Among the main *Meloidogyne* species occurring in Brazil, *M. incognita* (Kofoid & White, 1919), *M. arenaria* (Neal, 1889) and *M. javanica* (Treub, 1885) are the most frequent and widely studied (Luquini et al. 2019). These nematodes are responsible for substantial yield losses in several crops in the country, including tomato (*Solanum lycopersicum* L.), soybean (*Glycine max* (L.) Merr.), cotton (*Gossypium hirsutum* L.), common bean (*Phaseolus vulgaris* L.), corn (*Zea mays* L.) and coffee (*Coffea arabica* L.), among others (de Brida et al. 2020; Gonsaga et al. 2021; Balardin et al. 2022) and can also reproduce on weed species commonly found in cultivated areas (Bellé et al. 2019; Romagna et al. 2019; Kaspary et al. 2023). Although the host–parasite relationship in economically important crops is well documented, knowledge of this interaction in native plant species remains limited, representing a significant scientific gap.

Given the increasing relevance of native species and the need for their preservation, it is essential to assess their suitability within forest management, as they may influence reforestation programs (Santos et al. 2023; Lopes and Chiavari 2024). However, available information on the occurrence of potentially pathogenic nematodes in these trees is still limited, and such species may act as reservoirs of plant-parasitic nematodes of economic importance in Brazil. Historical evidence of the introduction of PPN associated with agricultural expansion highlights the need for preventive measures to ensure the sustainability of Brazilian agroecosystems (Márquez et al. 2021). In this context, the hypothesis that native tree species may multiply and even contribute to the dissemination of these nematodes motivated the present study, which aimed to evaluate the host status of 24 Brazilian native tree species to *M. incognita* and *M. javanica*.

2 | Material and Methods

Twenty-four native tree species were evaluated for their capacity to host and multiply two species of the genus *Meloidogyne* (*M. incognita* and *M. javanica*) (Table 1). The experiment was conducted in a greenhouse ($\approx 26^{\circ}\text{C}$, 80% RH) under a completely randomised design, with eight replicates per tree species for each nematode species. Thus, for every host species tested, eight plants were inoculated with *Meloidogyne incognita* and eight plants with *M. javanica*. Tomato (*Solanum lycopersicum* cv. Santa Clara) was used as a control for inoculum validation, whereas seedlings of the exotic tree *Tectona grandis* (Verbenaceae), known to promote the multiplication of *Meloidogyne* nematodes, served as a control reference.

For seedling production, seeds were sown and covered with a thin layer of autoclaved, moistened soil. The seedlings were then transplanted individually into plastic pots (4 dm^3 ; $200\text{ mm} \times 160\text{ mm}$) filled with an autoclaved sand-to-soil mixture (2:1). The soil used in the experiment, classified as a typical Oxisol (USDA-NRCS) or Rhodic Ferralsol (WRB), had the following physical–chemical properties: clay = 52%; pH (H_2O) = 6.0; organic matter = 2.8%; phosphorus = 10.1 mg dm^{-3} ;

potassium = 85 mg dm^{-3} ; calcium = $5.0\text{ cmol}_c\text{ dm}^{-3}$; magnesium = $4.7\text{ cmol}_c\text{ dm}^{-3}$; and sulfur = $9.0\text{ cmol}_c\text{ dm}^{-3}$.

Pure isolates of *M. javanica* and *M. incognita* were maintained and multiplied on tomato (*Solanum lycopersicum* cv. Santa Clara). The identification of *Meloidogyne* spp. isolates was confirmed by biochemical electrophoretic analysis of the non-specific esterase phenotype of females (Carneiro and Almeida 2001; Carneiro et al. 2016). Nematode inoculum was obtained from infected tomato roots following the methodology of Hussey and Barker (1973). Each plant was inoculated with a suspension containing 5000 eggs and second-stage juveniles (J2; initial population density, Pi), applied into three holes approximately 2 cm deep around the root system, 10 days after transplantation.

Tree species (including the *T. grandis* control) were uprooted 270 days after inoculation with *Meloidogyne*, whereas the nematode viability control (tomato) was uprooted 90 days after inoculation. Root systems were washed under running water, after which the number of galls was recorded and the gall index determined according to Taylor and Sasser (1978), where 0 = no galls, 1 = 1–2, 2 = 3–10, 3 = 11–30, 4 = 31–100 and 5 = more than 100 galls per root system. Eggs and second-stage juveniles (J2) were then extracted from the roots of each plant (final population density, Pf) following the methodology of Hussey and Barker (1973), and the nematode reproduction factor ($\text{RF} = \text{Pf}/\text{Pi}$) was calculated. Plants were classified according to their RF values as immune (I; $\text{RF} = 0$), resistant (R; $\text{RF} < 1.0$) or susceptible (S; $\text{RF} > 1.0$) (Oostenbrink 1966). In addition, the number of nematodes per gram of root was estimated as the ratio between the total number of nematodes and the total root mass (g) of each replicate.

Data were initially evaluated for normality using the Shapiro–Wilk test. Visual inspection of the frequency distributions (Figure S1) indicated strong deviations from normality, which were confirmed by the Shapiro–Wilk test ($p < 0.05$). Data transformation using $\sqrt{x+1}$ did not normalise the distributions. Therefore, all variables were analysed using non-parametric methods. Differences among species were assessed using the Kruskal–Wallis test, and when significant effects were detected, multiple comparisons were performed using Bonferroni-adjusted pairwise tests. All statistical analyses were conducted in R software.

3 | Results

For *M. incognita*, significant differences among species were detected for gall index (GI), nematodes per gram of roots (NNGR), and reproduction factor (RF) (Kruskal–Wallis test; GI: $H = 178.2$, $\text{df} = 24$, $p < 0.001$; NNGR: $H = 185.4$, $\text{df} = 24$, $p < 0.001$; RF: $H = 191.9$, $\text{df} = 24$, $p < 0.001$). Similarly, for *M. javanica*, significant differences among species were detected for GI ($H = 183.3$, $\text{df} = 24$, $p < 0.001$), NNGR ($H = 190.4$, $\text{df} = 24$, $p < 0.001$) and RF ($H = 194.1$, $\text{df} = 24$, $p < 0.001$).

For *M. javanica*, the highest gall index (GI) values were recorded in *Ilex paraguariensis* A. St.-Hil. (Aquifoliaceae), *Mimosa scabrella* Benth. and *Senna macranthera* (DC. ex Collad.) H.S.

TABLE 1 | Gall index (GI), number of nematodes per gram of roots (NNGR) and reproduction factor (RF) of *Meloidogyne javanica* (Treub, 1885) in native forest plants of Brazil grown under greenhouse conditions.

Family	Scientific name	GI		NNGR		RF	
Aquifoliaceae	<i>Ilex paraguariensis</i> A. St.-Hil.	5.0±0.00	a	758±172	cd	3.3±0.65	e
Bignoniaceae	<i>Handroanthus albus</i> (Cham.) Mattos.	4.9±0.35	a	1207±339	abc	5.9±1.26	bcd
	<i>Handroanthus impetiginosus</i> (Mart. ex DC.) Mattos.	4.4±0.52	abcd	917±574	cd	3.9±0.71	de
	<i>Handroanthus umbellatus</i> (Sond.) J.R.Mattos	3.9±0.35	cde	317±89	gh	1.9±0.45	fg
Fabaceae	<i>Anadenanthera falcata</i> (Benth.) Altschul	4.4±0.74	abc	469±112	efg	1.6±0.25	gh
	<i>Apuleia leiocarpa</i> (Vogel) J.F.Macbr.	0.0±0.00	j	0±0	l	0.0±0.00	l
	<i>Bauhinia forficata</i> Link.	4.6±0.52	ab	505±145	ef	2.1±0.49	f
	<i>Enterolobium contortisiliquum</i> (Vell.) Morong	4.6±0.52	ab	6029±1740	a	9.7±1.17	a
	<i>Erythrina falcata</i> Benth.	1.9±0.35	gh	83±29	jk	0.3±0.13	ij
	<i>Inga edulis</i> Mart.	0.3±0.46	ij	18±9	kl	0.1±0.03	k
	<i>Mimosa scabrella</i> Benth.	5.0±0.00	a	912±160	bcd	4.7±0.69	cde
	<i>Parapiptadenia rigida</i> (Benth.) Brenan	1.0±0.00	hij	26±14	jk	0.1±0.08	jk
	<i>Peltophorum dubium</i> (Spreng.) Taub.	3.1±0.35	efg	494±121	ef	1.6±0.30	gh
	<i>Senna macranthera</i> (DC. ex Collad.) H.S.Irwin and Barneby	5.0±0.00	a	1734±470	ab	6.2±1.04	abc
Moraceae	<i>Ficus adhatodifolia</i> Schott	3.6±0.52	de	671±64	hi	2.2±0.20	f
Malvaceae	<i>Luehea divaricata</i> Mart and Zucc.	3.5±1.30	bcde	330±173	de	1.5±0.40	f
Meliaceae	<i>Cedrela fissilis</i> Vell.	3.6±0.93	def	263±48	fgh	2.0±0.23	gh
Myrtaceae	<i>Campomanesia guazumifolia</i> (Cambess.) O. Berg	1.5±0.76	hi	59±31	jk	0.3±0.13	ijk
	<i>C. xanthocarpa</i> (O.Berg)	0.0±0.00	j	0±0	l	0.0±0.00	l
	<i>Eugenia involucrata</i> DC.	4.0±0.00	bcde	282±98	h	1.3±0.28	h
	<i>E. pyriformis</i> Cambess.	0.0±0.00	j	0±0	l	0.0±0.00	l
	<i>E. uniflora</i> L.	2.3±0.46	fgh	257±170	hi	0.5±0.24	i
	<i>Myrcianthes pungens</i> (O.Berg) D. Legrand	0.0±0.00	j	0±0	l	0.0±0.00	l
Sapindaceae	<i>Cupania vernalis</i> Cambess.	1.0±0.00	hij	104±37	ij	0.5±0.12	i
Lamiaceae	<i>Tectona grandis</i> L. f.	5.0±0.33	a	1387±535	ab	9.0±3.09	ab
Solanaceae	<i>Solanum lycopersicum</i>	5.0±0.0		5590±1200		39.8±5.02	
CV (%)		28.9		86.4		40.2	

Note: GI (gall index): 0=no gall, 1=1 to 2, 2=3 to 10, 3=11 to 30, 4=31 to 100 and 5=more than 100 galls per system root. NNGR (number of nematodes per gram of root): total number of nematodes/total root mass. RF (reproduction factor): final population density/initial population density. HS (host status): immune (I) (RF=0), resistant (R) (RF <1.0) and susceptible (S) (RF ≥ 1.0). Means followed by the same letter in each column do not differ significantly according to Bonferroni-adjusted pairwise comparisons following the Kruskal–Wallis test. Bignoniaceae nomenclature follows Grose and Olmstead (2007).

Irwin & Barneby (Fabaceae), *Handroanthus albus* (Cham.) Mattos (Bignoniaceae) and *Tectona grandis* L.f. (Lamiaceae; positive control) (Table 1). For *M. incognita*, the highest GI values were observed in *Enterolobium contortisiliquum* (Vell.) Morong, *M. scabrella* and *S. macranthera* (Fabaceae), as well as *H. albus* and *H. impetiginosus* (Mart. ex DC.) Mattos (Bignoniaceae) (Table 2). Nematodes per gram of roots (NNGR) ranged from 0 to 6029 for *M. javanica*, with the highest value recorded in *E. contortisiliquum*, which was 3.4-fold higher than that observed in *S. macranthera* (1734), the second-ranked species. Three species showed no nematode counts in their roots: *Apuleia leiocarpa* (Vogel) J.F. Macbr. (Fabaceae),

Campomanesia xanthocarpa (O. Berg) and *Myrcianthes pungens* (O. Berg) D. Legrand. For *M. incognita*, NNGR ranged from 0 to 7266. *A. leiocarpa* and *M. pungens* showed no nematodes in their roots, whereas *E. contortisiliquum* exhibited the highest NNGR value (7266) (Tables 1 and 2).

The reproduction factor (RF) of *M. javanica* ranged from 0 to 9.7, and that of *M. incognita* from 0 to 14.7. The highest RF values for both species were observed in *E. contortisiliquum* (Fabaceae), followed by *H. albus* (Bignoniaceae) and *S. macranthera* (Fabaceae) for both nematodes, while *Cedrela fissilis* Vell.

TABLE 2 | Gall index (GI), number of nematodes per gram of roots (NNGR) and reproduction factor (RF) of *Meloidogyne incognita* (Kofoid & White, 1919) in native forest plants of Brazil grown under greenhouse conditions.

Family	Scientific name	GI		NNGR		RF	
Aquifoliaceae	<i>Ilex paraguariensis</i> A. St.-Hil.	3.5 ± 0.53	efg	913 ± 208	cde	5.0 ± 0.98	ef
Bignoniaceae	<i>Handroanthus albus</i> (Cham.) Mattos.	5.0 ± 0.00	a	1455 ± 408	abcd	8.8 ± 1.90	bcd
	<i>Handroanthus impetiginosus</i> (Mart. ex DC.) Mattos.	5.0 ± 0.00	a	1105 ± 691	cde	5.9 ± 1.07	de
	<i>Handroanthus umbellatus</i> (Sond.) J.R.Mattos	4.4 ± 0.52	abcd	382 ± 107	hij	2.9 ± 0.67	gh
Fabaceae	<i>Anadenanthera falcata</i> (Benth.) Altschul	4.3 ± 0.46	bcde	566 ± 135	gh	2.4 ± 0.38	hi
	<i>Apuleia leiocarpa</i> (Vogel) J. F. Macbr.	0.0 ± 0.0	j	0 ± 0	n	0.0 ± 0.00	m
	<i>Bauhinia forficata</i> Link.	4.0 ± 0.00	cdef	609 ± 175	fgh	3.2 ± 0.73	fgh
	<i>Enterolobium contortisiliquum</i> (Vell.) Morong	5.0 ± 0.00	a	7266 ± 2109	a	14.7 ± 1.77	a
	<i>Erythrina falcata</i> Benth.	1.8 ± 0.46	hij	100 ± 35	klm	0.5 ± 0.21	kl
	<i>Inga edulis</i> Mart.	1.1 ± 0.99	ij	22 ± 11	mn	0.1 ± 0.07	lm
	<i>Mimosa scabrella</i> Benth.	5.0 ± 0.00	a	1099 ± 193	bcde	7.1 ± 1.04	cde
	<i>Parapiptadenia rigida</i> (Benth.) Brenan	1.9 ± 0.99	hi	32 ± 17	mn	0.2 ± 0.12	lm
	<i>Peltophorum dubium</i> (Spreng.) Taub.	4.5 ± 0.53	abc	596 ± 146	fgh	2.5 ± 0.46	ghi
	<i>Senna macranthera</i> (DC. ex Collad.) H.S.Irwin and Barneby	5.0 ± 0.00	a	2090 ± 566	ab	9.3 ± 1.58	abc
	<i>Ficus adhatodifolia</i> Schott	4.9 ± 0.35	ab	903 ± 266	def	9.0 ± 3.0	bcd
Malvaceae	<i>Luehea divaricata</i> Mart and Zucc.	3.6 ± 0.83	cdef	809 ± 208	efg	2.3 ± 0.59	fg
Meliaceae	<i>Cedrela fissilis</i> Vell.	4.9 ± 0.52	defg	903 ± 58	hij	9.0 ± 0.34	hi
Myrtaceae	<i>Campomanesia guazumifolia</i> (Cambess.) O. Berg	1.4 ± 0.52	ij	71 ± 37	lm	0.4 ± 0.18	kl
	<i>C. xanthocarpa</i> (O.Berg)	1.4 ± 0.52	ij	63 ± 40	lmn	0.4 ± 0.25	l
	<i>Eugenia involucrata</i> DC.	3.9 ± 1.25	cdefg	339 ± 254	ijk	1.9 ± 0.42	ij
	<i>E. pyriformis</i> Cambess.	3.5 ± 0.53	efg	601 ± 192	fgh	2.0 ± 0.60	ij
	<i>E. uniflora</i> L.	3.9 ± 1.25	fgh	339 ± 254	ghi	1.9 ± 0.42	jk
	<i>Myrcianthes pungens</i> (O.Berg) D. Legrand	0.0 ± 0.0	j	0 ± 0	n	0.0 ± 0.0	m
Sapindaceae	<i>Cupania vernalis</i> Cambess.	2.6 ± 1.19	ghi	257 ± 84	jkl	1.5 ± 0.45	j
Lamiaceae	<i>Tectona grandis</i> L. f.	5.0 ± 0.33	a	1655 ± 645	abc	13.6 ± 4.65	ab
Solanaceae	<i>Solanum lycopersicum</i>	5.0 ± 0.0		6740 ± 1900		60.6 ± 9.22	
CV (%)		23.0		75.1		43.4	

Note: GI (gall index): 0=no gall, 1=1 to 2, 2=3 to 10, 3=11 to 30, 4=31 to 100 and 5=more than 100 galls per system root. NNGR (number of nematodes per gram of root): total number of nematodes/total root mass. RF (reproduction factor): final population density/initial population density. HS (host status): immune (I) (RF=0), resistant (R) (RF < 1.0) and susceptible (S) (RF ≥ 1.0). Means followed by the same letter in each column do not differ significantly according to Bonferroni-adjusted pairwise comparisons following the Kruskal–Wallis test. Bignoniaceae nomenclature follows Grose and Olmstead (2007).

(Meliaceae) showed a high RF only for *M. incognita*. Considering the total of 24 tree species evaluated, 62.5% were classified as susceptible to *M. javanica*, whereas 70.8% were susceptible to *M. incognita*. Six species were resistant (RF < 1.0) to *M. javanica* and five to *M. incognita*, with four resistant to both: *Campomanesia guazumifolia* (Cambess.) O. Berg, *Erythrina falcata* Benth., *Inga edulis* Mart. and *Parapiptadenia rigida* (Benth.) (Table S1). *Campomanesia xanthocarpa* (O.Berg) resisted only *M. incognita*, whereas *Casearia vernalis* Cambess. and *Eugenia uniflora* L. resisted only *M. javanica*. Four species were immune to *M.*

javanica (*A. leiocarpa* and three Myrtaceae: *C. xanthocarpa*, *E. pyriformis* Cambess. and *M. pungens*), and two (*A. leiocarpa* and *M. pungens*) were immune to *M. incognita*; these latter two were the only species immune to both root-knot nematodes.

4 | Discussion

The results provide evidence that *M. incognita* and *M. javanica* can parasitize and reproduce in some Brazilian native tree species.

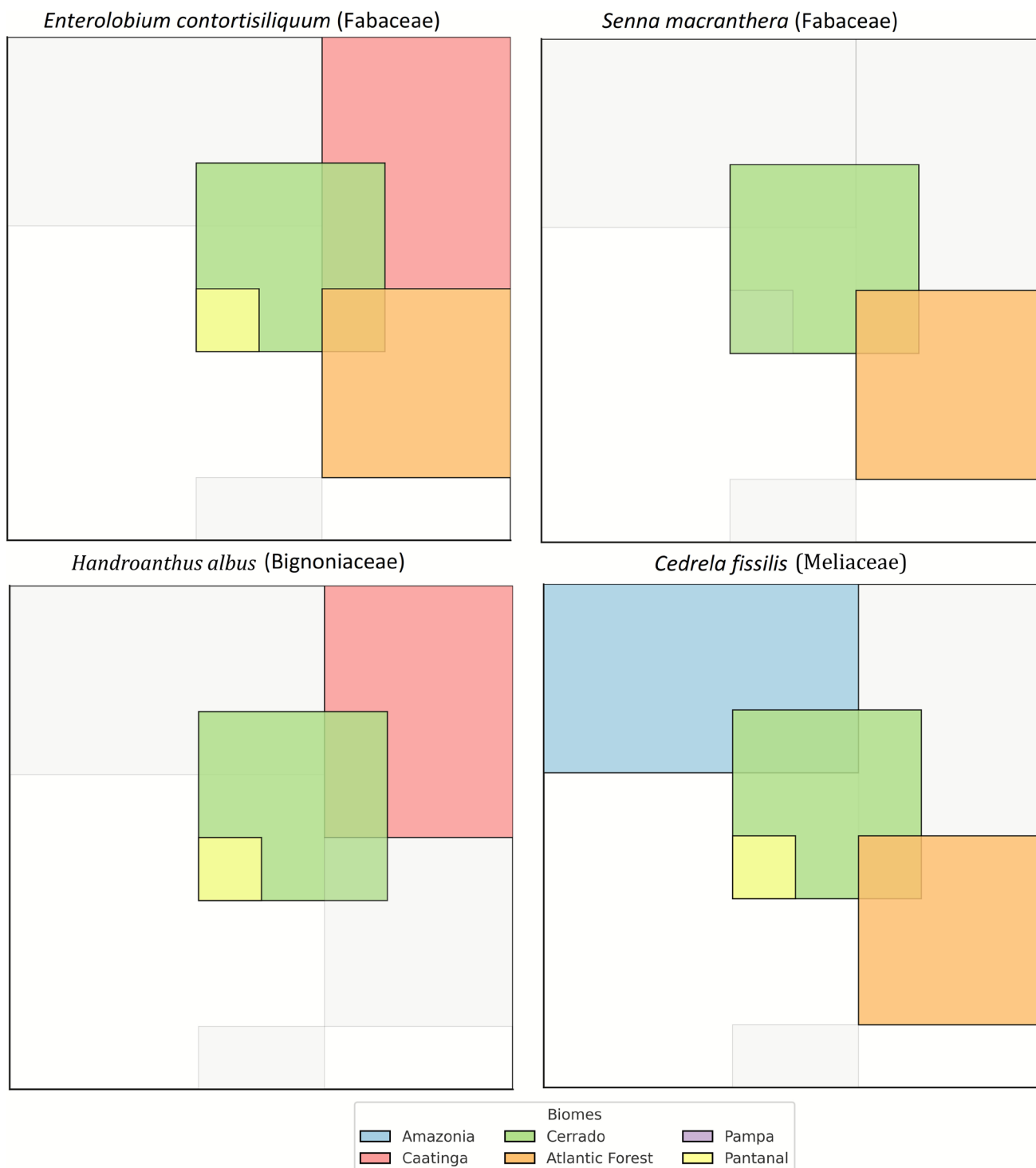


FIGURE 1 | Estimated distribution of selected native tree species across Brazilian biomes. Each map highlights the natural occurrence of *Enterolobium contortisiliquum* (Vell.) Morong (Fabaceae), *Senna macranthera* (DC. ex Collad.) H.S.Irwin & Barneby (Fabaceae), *Cedrela fissilis* Vell. (Meliaceae) and *Handroanthus albus* (Cham.) Mattos. (Bignoniaceae). Colours represent the six major Brazilian biomes: Amazonia (blue), Caatinga (red), Cerrado (green), Atlantic Forest (orange), Pampa (purple) and Pantanal (yellow). Grey areas indicate absence of records for the species. Distribution data derived from Flora do Brasil 2020.

However, with few exceptions, most exhibited low RF compared with the control (*T. grandis*). Overall, 41.6% (10) and 29.2% (7) of the tested species were resistant or immune to *M. javanica* and *M. incognita*, respectively. In a study from the Brazilian Cerrado, 80% (12 species) were immune to *M. javanica* (Silva et al. 2014), and five RKN species (*M. arenaria*, *M. hapla* Chitwood, *M. incognita*,

M. javanica and *M. morocciensis* Rammah & Hirschmann) were detected in 22.8% of root samples from native trees.

The high reproduction factor of *M. javanica* (RF=9.7) and *M. incognita* (RF=14.7) in the roots of *E. contortisiliquum* (Fabaceae) evidence that this tree is a highly favourable host for

these nematodes. Although the underlying mechanisms were not investigated in this study, the high susceptibility observed in several Fabaceae species may be associated with root architectural traits and chemical signalling that favour nematode penetration and reproduction, as suggested for other plant–nematode interactions (Perry et al. 2024). Widely distributed in the Atlantic Forest, Cerrado and Caatinga biomes (Figure 1), this species is also used in reforestation programs due to its rapid growth and adaptability to different edaphoclimatic conditions (Naves et al. 2018). Under natural conditions, *E. contortisiliquum* may contribute to the maintenance and dissemination of *M. javanica* and *M. incognita*. This host potential is further supported by the fact that *M. enterolobii* (Yang & Eisenback, 1983)—one of the most aggressive root-knot nematodes—was originally described from a population associated with *E. contortisiliquum* (Damascena et al. 2019). It is also noteworthy that *S. macranthera* (Fabaceae), *H. albus* (Bignoniaceae) and *C. fissilis* (Meliaceae), which also showed high RF values, share a common distribution in the Cerrado biome (Figure 1). As the Cerrado is the main region for grain, fibre and cellulose production in Brazil, these native species may serve as refuges for nematodes in areas adjacent to agricultural fields and commercial forests.

Previous studies reinforce the potential of native plants as hosts for RKN. In the Atlantic Forest, *Meloidogyne* spp. were detected in 21 of 360 soil samples, with *M. javanica* and *M. exigua* being the most abundant (Lima et al. 2005). In another study, *M. incognita* and *M. javanica* occurred naturally in 11.2% and 8.3% of the evaluated native species, respectively; moreover, when 29 species were inoculated, *M. incognita* multiplied in only 10.3% of them (Antes et al. 2012). Native trees have also been reported as hosts for other plant-parasitic nematodes. Favoreto et al. (2013) found that *Inga uruguensis* (Fabaceae), *Handroanthus albus* and *Tabebuia heptaphylla* (Bignoniaceae) acted as hosts for *Pratylenchus brachyurus*. Similarly, Silva et al. (2008) identified the genera *Xiphinema*, *Meloidogyne*, *Pratylenchus*, *Helicotylenchus* and *Rotylenchus* in preserved areas of the Amazon rainforest.

Several native tree species susceptible to *M. incognita* and *M. javanica*, identified in this study, are common in forest edges adjacent to agricultural fields. These species may act as alternative hosts and inoculum reservoirs for nearby areas or contribute to a silent decline of native plants in secondary vegetation. For example, in Spain, *M. silvestris* was found causing high infection rates in the roots of European holly (*Ilex aquifolium*), being associated with forest decline (Castillo et al. 2009). Another risk is the use of non-sterilised soil in nurseries, which may introduce nematodes into seedlings intended for landscaping, ecological restoration or native fruit production. In Morocco, Hamza et al. (2017) recorded a high incidence of *M. javanica* in nurseries and its subsequent detection in orchards, suggesting introduction through seedlings. Similarly, in Brazil, Antes et al. (2012) detected *M. incognita* and *M. javanica* in the roots of seven out of 36 native tree species evaluated in a commercial nursery in the south of the country.

To date, the economic damage caused by plant-parasitic nematodes to native plants in natural areas and secondary vegetation remains unknown. The occurrence, diversity and distribution

of these organisms in Brazilian biomes still represent a major knowledge gap, supported only by sporadic studies in native or reforested areas. This gap highlights the need for further research, especially because many native tree species that host nematodes have economic value and are widely used in landscaping, reforestation, agroforestry systems, rustic timber production and the extraction of compounds for phytotherapeutics. Moreover, native species found to be immune to the evaluated nematodes may harbour resistance genes or bioactive compounds of biotechnological interest, offering new opportunities for nematode management and sustainable control strategies. From a practical perspective, the identification of native tree species that are susceptible to root-knot nematodes has direct implications for forest restoration and nursery sanitation, as these species may act as inoculum reservoirs when propagated in non-sterilised substrates. Conversely, resistant or immune species may represent safer alternatives for reforestation programs in nematode-infested areas.

Author Contributions

Ismail Teodoro de Souza Júnior, Tiago Edu Kaspary and Cristiano Bellé contributed to the study conception and design, experimental setup and data collection. Rodrigo Ferraz Ramos and Zaida Inês Antonioli performed the data analysis and led the final manuscript writing. Vicente Guilherme Handte and Juliano Borela Magalhães contributed to the initial draft and manuscript editing.

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Conflicts of Interest

The authors declare no conflicts of interest.

Data Availability Statement

The data that support the findings of this study are available in the [Supporting Information](#) of this article.

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Supporting Information

Additional supporting information can be found online in the Supporting Information section. **Data S1:** Raw data provided in a Microsoft Excel spreadsheet (.xlsx). The file contains gall index, number of nematodes per gram of root and reproduction factors for *M. incognita* and *M. javanica*. **Figure S1:** Frequency distributions and exploratory assessment of data structure for gall index (GI), nematodes per gram of roots (NNGR) and reproduction factor (RF) in native tree species inoculated with *Meloidogyne incognita* (Kofoed & White, 1919) and *M. javanica* (Treub, 1885) under greenhouse conditions. (A) Frequency distribution of gall index (GI; scores 0–5) for *M. incognita* and *M. javanica*. (B) Theoretical standard normal distribution (mean = 0, SD = 1) shown for visual comparison. (C, D) Histograms of NNGR values for *M. incognita* (C) and *M. javanica* (D), displayed on a density scale. (E, F) Histograms of reproduction factor (RF) values for *M. incognita* (E) and *M. javanica* (F), displayed on a density scale. In panels C–F, dashed red lines represent normal distributions fitted using the observed mean and standard deviation. **Table S1:** Host response of native Brazilian tree species to *Meloidogyne incognita* and *M. javanica* under greenhouse conditions.