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Nutrient Omission and *Aphelenchoides besseyi* Interaction in Soybean Plants

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ABSTRACT

The phytonematode *Aphelenchoides besseyi* gained prominence in Brazil following its identification as the etiological agent of leaf and green stem retention in soybean plants. Thus, this study evaluated the effects of nutrient omission on the phytotechnical characteristics of soybean in the presence and absence of *A. besseyi*. The experiments were carried out in a completely randomized 2 × 11 factorial arrangement, with 11 treatments: control (no nutrients), complete nutrition, and individual -N, -P, -K, -S, -B, -Cu, -Fe, -Mn, and -Zn omission were applied to inoculated and uninoculated plants, and six replicates. The presence of *A. besseyi* adversely affected the growth characteristics, except when -P, -S, -B, -Mn, and -Zn were omitted. The nematode reduced the root length in treatments omitting -K, -S, -B, -Cu, -Fe, -Mn, and -Zn and root fresh weight when -N, -P, -K, and -Cu were omitted. In the absence of the pathogen, N assimilation did not differ significantly between the treatments; however, it was negatively influenced by the presence of the pathogen in the control and complete nutrition treatment. The leaf area index values were the lowest in inoculated plants with no nutrition, with complete nutrition, and with -N, -P, -K, and -Cu omissions. The micro- and macronutrients concentration and the shoot dry weight (SDW) were high in the presence of the nematode, except in treatments where -N, -S, and -Mn were omitted. The nutritional status does not affect infection by the nematode *A. besseyi*, but the application of nutrients is necessary for soybean development.

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Agronomic characteristics; *Glycine max*; green stem nematode; nutritional status

Introduction

Aphelenchoides besseyi is a phytonematode that preferentially attacks the shoot of plants (Meyer et al. 2017), and it has been gaining interest in the Brazilian agricultural scenario owing to its high potential to damage to crops such as soybean (*Glycine max* [L.] Merrill) and cotton (*Gossypium hirsutum* L.). The ability to enter anhydrobiosis and survive for prolonged periods in unfavorable environments allows this organism to persist for a long time in the field (Favoretto and Meyer 2019).

Rice (*Oryza sativa* L.) is the primary host of *A. besseyi* throughout the world, wherein it causes the "white tip" disease (Kepenekci 2013). However, in Brazil, this nematode has gained significance when it was identified as the etiological agent of leaf retention and green stem in soybean plants (Meyer et al. 2017). Using integrated measures such as biological and chemical control and crop succession, the nematode population has been efficiently controlled, and the incidence of the disease has been reduced (Favoretto and Meyer 2019).

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The most characteristic symptoms of *A. besseyi* are observed in the soybean plants, as evidenced by the deformed stems and thickened nodes, thinned and blistered leaf blades, thickened veins of young leaves, abortion of flowers and pods, possibly with super budding; the remaining viable pods are deformed and frequently possess brown necrotic lesions (Meyer et al. 2017).

Despite the increased infestation in the tropics, the effects of the nutritional status of the plant on the parasitic relationships of *A. besseyi* are not yet examined even though nutrient deficiency is known to directly or indirectly affect the host – pathogen interaction, alter the soil environment, and promote or reduce the presence of pathogens, thus, affecting the severity of the disease, as well as inducing resistance or tolerance in the host plant (Almeida and Seixas 2010; Dias et al. 2021; Zambolim, Costa, and F 2001). Nutritional requirements correspond to the total quantity of nutrients that are absorbed and are necessary for the vegetative and reproductive development of plants. Nutrients have essential and specific functions in plants, and their deficiency or excess can trigger changes in plant metabolism that influence crop productivity (Malavolta 2006; Marschner 2012).

The nematodes infest the leaves, causing a reduction in size and yellowing of the plants, and similar symptoms are caused by a lack of N, S, Mo, and Fe. In some cases, the leaves show chlorotic spots and necrosis between veins, which resemble symptoms of K, Mg, Cu, and Mn deficiency. Additionally, pod abortion is also associated with B and Ca deficiencies (Dias et al. 2010; Malavolta, Vitti, and Oliveira 1997).

The objective of this study was to evaluate the influence of nutrient omission on the phytotechnical characteristics, chlorophyll content, and leaf area index (LAI) in the soybean plants cultivated in the presence and absence of *A. besseyi*.

Materials and methods

The experiments were conducted in the greenhouse at Embrapa Soja with an average temperature of 24°C (±2 °C) and constant spraying for 15 s every 20 min. The experimental design was a completely randomized 2 × 11 factorial arrangement with 11 treatments: control (without nutrients), complete nutrition, and nitrogen (N), phosphorus (P), potassium (K), sulfur (S), boron (B), copper (Cu), iron (Fe), manganese (Mn), and zinc (Zn) individual omission, for uninoculated and *A. besseyi*-inoculated plants, with six replicates per treatment.

The soil used as part of the substrate was typical Red Oxisol collected at 0.0–0.2 m depth in the Ponta Grossa County, Paraná State, Brazil having the following chemical attributes (Embrapa (Empresa Brasileira de Pesquisa Agropecuária) 1997): pH ($CaCl_2$) = 4.8, soil organic matter (SOM) = 51.3 g kg^{-1}, P (Mehlich 1) = 5.3 mg kg^{-1}, Ca^{2+} = 3.1 $cmol_c$ kg^{-1}, Mg^{2+} = 2.3 $cmol_c$ kg^{-1}, K+ = 0.4 $cmol_c$ kg^{-1}, Al^{3+} = 0.4 $cmol_c$ kg^{-1}, potential acidity (H+Al) = 6.4 $cmol_c$ kg^{-1}, cation exchange capacity (CEC) = 12.2 $cmol_c$ kg^{-1}, clay = 600 g kg^{-1}, and sand = 170 g kg^{-1}.

The soybean cultivar BRS 284, which is susceptible to *A. besseyi* was used for the experiments. The soil was autoclaved to eliminate the presence of nematodes, mixed in a 3:1 ratio of sand: clay, and placed in 1.0 L pots. Two seeds were sown per pot, and thinning was performed 10 days after emergence, leaving only one plant. Next, 500 nematodes were inoculated per pot by placing their suspension in a hole approximately 2.0 cm deep in the soil, near the plant's neck.

The nematodes were obtained from a pure population of *A. besseyi*, multiplied in the dark for 30 days in Petri dishes with colonies of *Fusarium* sp. (approximately 5-day-old growth on potato dextrose agar medium) in BOD-type chambers at 25°C (±1°C) (Favoreto et al. 2011). The nematodes were collected in 25 µm sieves by washing the insides of the Petri dish lids with water to prepare the inoculum suspension in water. The uninoculated plants received water instead of the nematode suspension.

After 10 days of nematode inoculation, the nutrient solutions previously prepared as described in Moreira et al. (2011), were applied to the pots for the greenhouse experiments as per the specified treatments. The complete nutrient treatment solution contained N, P, K, S, B, Cu, Fe, Mn, and Zn, in the form of 100 mg kg^{-1} of N as CH_4N_2O (45% N), 150 mg kg^{-1} of P as $NH_4H_2PO_4$ (54% P_2O_5), 100 mg kg^{-1} of K as KCl (60% K_2O), 100 mg kg^{-1} of S as elemental S (98% S), 1.0 mg kg^{-1} of B as H_3BO_3 (18% B), 2.0 mg kg^{-1} of Cu as $CuCl_2{\cdot}2\,H_2O$, 2.5 mg kg^{-1} $FeCl_2$, 2.5 mg kg^{-1} of Mn as $MnCl_2{\cdot}H_2O$, and 5.0 mg kg^{-1} of Zn as $ZnCl_2$, while for the other treatment solutions one nutrient was omitted per treatment.

Nutrients were applied over 4 days, with an average of 2–3 applications per day. Plastic dishes were placed below each pot. When necessary, the water retained in the plastic dishes was returned to the pot to prevent the loss of nutrient solution. Evaluations were performed after 40 days of nutrient solution application and 50 days of nematode inoculation. Plant height (PH) was measured from the ground level to the apex of the main branch of the plant, with the aid of a ruler. The shoot fresh weight (SFW) was determined by weighing the shoots immediately after removal from the pot, and root fresh weight (RFW) was determined after washing the roots in running water to remove the adhered soil, and resting for 120 min. The root length (RL) was measured immediately after weighing. The relative chlorophyll content (SPAD index with the KONICA MINOLTA® SPAD 502 chlorophyll meter) was estimated by sampling the third trifoliate from the apex to the base of the plant and considering the central leaflet. LAI was measured using a leaf area integrator (LI-COR®, model LI 3100). The numbers of nodes (ND), flowers (NF), small pods (NP), and pods per plant (NPP) were also evaluated.

For nutrient estimation, the shoots and the third and fourth trifoliate of the plants were collected and ground into a paste and then subjected to chemical analysis to determine the main macronutrients (N, P, K, and S) and micronutrients (B, Cu, Fe, Mn, and Zn) using the methodologies described by Malavolta, Vitti, and Oliveira (1997). N was determined by titration; P, S, and B by colorimetry; K was quantified by flame photometry; and the total Cu, Fe, Mn, and Zn contents in plant tissue were determined by atomic absorption spectrophotometry.

The symptoms of deficiency and/or toxicity of each treatment were also evaluated during plant growth, and at the end of the trial, a comparison was made between the inoculated and uninoculated plants for each nutrient treatment. For nematological evaluation, the shoots were processed using the methodology described by Coolen and D´Herde (1972), and the nematodes were quantified in a Peters chamber under a light microscope at 100× magnification.

For the uninoculated plants, two additional phytotechnical characteristics were analyzed: shoot dry weight yield (SDWY), and root volume (RV). SDWY was estimated by drying the previously identified shoot samples in an oven. The RV was determined by the water displacement method, where a known volume of water was taken in a beaker, and the roots were then immersed in the beaker with water, the RV was estimated as the volume of water displaced in the beaker and expressed in cm^3.

For the data analysis of PH, SFW, and the number of nematodes (*A. besseyi*), 5% significance was applied. Data from nutritional analyses were submitted to normality tests (Shapiro – Wilk) and analysis of variance (ANOVA) was applied, and the mean values were compared using Tukey's test at 5% significance. For comparing the RL, RFW, SPAD index, and LAI in response to soybean nutritional status and the presence (+) and absence (-) of *A. besseyi*, the error normality tests were performed (Shapiro – Wilk), and subjected to ANOVA, F test, and Scott – Knott test at 5% significance.

Results and discussion

The presence of *A. besseyi* reduced the pH in treatments where -N, -P, and -Cu were omitted, and the control (without nutrients), whereas in the uninoculated plants, the reduction in pH was observed only in the treatment with -P omission (Table 3.1). Treatments omitting -P, -S, -B, -Mn, and -Zn did not show significant differences in the presence or absence of phytonematodes. Although N is the most important nutrient required by soybean (Bahry et al. 2013) and its deficiency is known to limit plant growth (Maia et al. 2014), a greater restrictive effect of -N omission on plant development was expected but was not observed in the uninoculated plants in our study. With -P omission, the plants showed lesser height and SFW yield, which is concurrent with the results of Prado, Franco, and Puga (2010) that P deficiency causes stunted plants and a blue-green color. -Cu omission reduced plant growth and caused wilting and chlorosis along the leaf margins, corroborating the results of Maia et al. (2014) and Moreira et al. (2022), who reported that in soybean crops, the lack of these nutrients significantly reduced the shoot volume of the plants. The SFW yield was less in the presence of *A. Besseyi* with -N, -P, -K, and -Cu omission, and control in comparison to that with complete nutrition (Table 1). In the absence of the nematode, only the control and -P and -Zn omission showed a statistically significant difference, whereas the differences were not significant with and without the pathogen in the treatments omitting -P, -S, -B, -Mn, and -Zn (Table 1). The adverse effects of the presence of nematodes in relation to the nutritional status of plants were also verified by Dias et al. (2021), who observed a significant reduction in SFW yield when -N was omitted, and by Barreto et al. (2017), who reported a reduction in SFW yield of strawberry (*Fragaria* × *ananassa*) in treatments with -N, -P, and -K omission. Moreira et al. (2000), Alves et al. (2002), and Carlos et al. (2014) also showed that the nutritional disorder in plants alters their growth, development, and specific morphological characteristics, as verified with the Zn treatment, whose deficiency is reported to alter the formation of indole acetic acid, causing narrower leaves and short internodes (Moreira, Moraes, and Reis 2018). Unlike the results of the study on *M. javanica* by Dias et al. (2021), in which a negative effect on SFW yield was noted because of the low Zn content of the soil, -Zn omission in the present study did not cause chlorosis or a reduction of SFW yield (Table 1). The number of *A. besseyi* found in the SFW differed statistically between the treatments, with the highest yield noted in the complete treatment (Table 1). It stands to reason that even with the highest pathogen numbers in the shoot of the plant (Meyer et al. 2017), the supply of balanced essential nutrients can minimize the severity of the disease (Ferraz et al. 2012). The RL of soybean plants in presence of the nematode was less in the treatments with -K, -S, -B, -Cu, -Fe, -Mn, and -Zn omission, and the values differed significantly from the control, complete nutrition, and -N and -P omission (Table 2), whereas in the absence of the pathogen, such difference was not observed, and further comparison between the presence and absence of the nematode within each treatment showed that the -K, -S, -B, -Cu, -Fe, and -Mn omission did not affect RL significantly (Table 2).

Fageria and Moreira (2011) reported that RL is directly related to SFW and it positively influences the grain yield. Symptoms of nutritional deficiency in plants include metabolic disorders resulting from an insufficient supply of one or more elements, which are related to the specific functions of each nutrient in the metabolism and development of plants (Malavolta 2006; Marschner 2012).

In the presence of *A. besseyi*, RFW was the lowest in treatments with the -N, -P, -K, -Cu, and -Zn omission (Table 2), median with the -S, -B, -Fe, and -Mn omission, while the control and plants with

Table 1. Growth components [plant height (PH), shoot fresh weight (SFW)] of soybean in response to presence (+) and absence (-) and number of *Aphelenchoides besseyi* individuals.

	PH		SFW		Total *A. besseyi* in SFW
	(cm)		(g)		(n)
Treatments	(+)	(-)	(+)	(-)	(+)
Control	39.1 bB	85.3 aA	3.9 bA	4,3 bA	317b
Complete	56.1 aB	101.3 aA	4.8 aB	6,8 aA	527a
− N	42.3 bB	99.2 aA	4.3 bB	6,5 aA	263b
− P	39.3 bA	47.8 bA	3.9 bA	4,5 bA	200b
− K	56.7 aB	88.3 aA	4.7 bB	6,1 aA	230b
− S	69.0 aA	88.8 aA	5.5 aA	6,1 aA	297b
− B	64.0 aA	80.7 aA	5.0 aA	6,1 aA	238b
− Cu	39.5 bB	91.0 aA	4.2 bB	6,4 aA	245b
− Fe	63.5 aB	90.5 aA	5.3 aB	6,4 aA	343b
− Mn	58.5 aA	77.8 aA	5.1 aA	5,8 aA	312b
− Zn	65.5 aA	83.3 aA	4.9 aA	5,3 bA	217b
Mean	53.9B	84.9A	4.7B	5,8A	289,9
CV (%)	27.4%		16.8%		12.7%

Note: *Means followed by the same lowercase letters in columns and uppercase letters in rows do not differ by the Scott-Knott test ($p \leq .05$); ns = not significant ($p \leq .05$).

Table 2. Root length (RL), chlorophyll content, and leaf area index and root fresh weight (RFW) of soybean in response to the presence (+) and absence (-) of *Aphelenchoides besseyi*.

	RL		RFW		Chlorophyll		Foliar area	
	(+)	(-)	(+)	(-)	(+)	(-)	(+)	(-)
Treatments	(cm)		(g per pot)		(mg m^2)		(cm^2)	
Control	42.8aA	27.2aB	2.0bB	4.8 bA	263.1 bA	270.6aA	91.9bB	864.3 bA
Complete	39.8aA	27.3aB	3.0aB	5.6aA	346.4bB	283.4aA	159.4bB	1080.3aA
− N	44.0aA	20.2aB	1.1cB	4.8 bA	302.5aA	273.7aA	118.9bB	704.4cA
− P	43.3aA	20.8aB	0.9cB	1.8eA	394.1aA	246.4aB	118.3bB	202.5 dA
− K	28.3 bA	25.7aA	0.7cB	4.1cA	297.9aA	272.1aA	236.9aB	656.2cA
− S	28.3 bA	23.5aA	1.6bB	4.0cA	393.4aA	270.0aA	234.3aB	741.9cA
− B	24.0 bA	22.0aA	1.4bB	2.9 dA	297.9aA	256.2aB	232.6aB	683.4cA
− Cu	25.2 bA	21.3aA	0.9cB	4.7 bA	321.4aA	276.7aB	154.0bB	716.3cA
− Fe	25.8 bA	22.0aA	1.8bB	5.8aA	305.5aA	379.7aA	181.1aB	758.3cA
− Mn	19.3 bA	19.8aA	1.7bB	4.1cA	325.9aA	379.6aB	253.1aB	677.3cA
− Zn	32.7 bA	16.7aB	2.5aB	5.2aA	292.6aA	33.2aA	211.1aA	204.1 dA
Mean	32.1	22.4	1.5	4.3	294.9	270.6	181.0	662.6
F-Test								
Nutrients (a)	NS		*		*		NS	
Nematode (b)	*		*		*		*	
a × b	NS		*		*		*	
CV (%)	25.4		16.0		9.5		13.0	

Note: *Means followed by the same lowercase letters in columns and uppercase letters in rows do not differ by the Scott-Knott test ($p \leq .05$); * = significant and ns = not significant ($p \leq .05$).

complete nutrition showed the highest RFW. In treatments without nematode inoculation, the highest values of RFW were found in the plants with complete nutrition, and -Fe and -Zn omission (Table 2). P omission caused the lowest RFW, followed by the -B omission treatment. The treatments with -N, -K, -S, -Cu, and -Mn omission and without nutrition were statistically different and were in a median position among the other values. Similarly, Dias et al. (2021) reported that the absence of nematodes in all treatments resulted in significantly different values, with the lowest RFW values observed in the presence of the pathogen.

The chlorophyll content showed an average reduction of 9.0% in the treatments in the presence of *A. besseyi* in soybean (Table 2). Comparing the treatments with and without nematode infection, the infected plants showed a significant chlorophyll reduction with the values for -P, -B, and -Cu omission reduced by 37.5%, 14.0%, and 13.9%, respectively, while the -Mn omission treatment showed an increase of 16.5%. In the infected plants, the control and -B omission had the lowest values and were statistically different, and in the uninoculated plants, there was no difference in the chlorophyll content between the various treatments (Table 2). These results demonstrate that *A. Besseyi* infection in soybean causes physiological changes in the assimilation of nutrients that act directly or indirectly on energy generation and affect the plant metabolism, resulting in nutritional deficiencies or excesses, which can also characterize the symptoms of this endoparasite.

In the nematode infected plants, the control, complete nutrition, -N, -P, -K, and -Cu omission treatments showed the lowest values of LA and were significantly different from the other treatments (Table 2). For plants without *A. Besseyi*, the highest LA was found in the treatment with complete nutrition, while the treatments with -N, -K, -S, -B, -Cu, -Fe, and -Mn omission presented median values of LA, with the lowest values found in the treatments with -P and -Zn omission and this effect is in agreement with Marschner (2012) since these two nutrients (P and Zn) are directly related to shoot growth and their deficiency severely affects the growth.

RV, SDW, ND, NF, NP, and NPP were evaluated when the soybean plants were at the R2–R3 growth stage (Fehr et al. 1971) in the treatments without the pathogen, whereas in the presence of the pathogen and owing to the removal of the plants for evaluation, only the nodes were observed, and

Table 3. Root volume, shoot dry weight, number of nodes, number of flowers, small pods, and pods of plants without inoculation.

Treatments	RV (cm^3)	SDW (g)	Nós (n)	Flowers (n)	Small pods (n)	Pods (n)
Control	8.3b	5.2a	17.3a	1.3c	2.8c	6.0c
Complete	13.3a	7.8a	23.5a	20.2a	7.0a	11.5a
– N	7.1c	7.2a	20.2a	3.4c	4.5b	7.5b
– P	4.3c	1.9b	9.0b	9.0b	2.3c	4.7c
– K	3.4c	6.0a	19.3a	19.3a	5.5a	6.0c
– S	4.7c	5.8a	19.0a	18.5a	4.2b	8.0b
– B	3.1c	5.7a	19.7a	17.3a	4.3b	9.5a
– Cu	3.3c	6.3a	18.7a	18.7a	4.5b	11.0a
– Fe	4.9c	6.5a	15.7b	15.7b	5.2a	9.2a
– Mn	3.5c	6.1a	13.5b	13.5b	3.7b	7.3b
– Zn	4.9c	4.7a	12.7b	12.7b	1.8c	5.2c
Mean	5.5	5.7	17.1	13.6	4.2	7.8
CV%	24.6	21.1	19.5	22.3	19.4	18.8

Note: *Means followed by the same lowercase letters in columns do not differ by the Scott-Knott test ($p \leq .05$).

some of them were necrotic (Table 3). According to Fageria and Moreira (2011), the results show the importance of balanced fertilization, as the highest RV was obtained with complete nutrient treatment, differing statistically from the other treatments (Table 3). For SDW, the -P omission treatment had the lowest value, with a reduction of 278.9% compared to that of the complete treatment (Table 3). Regarding the NF, ND, NPP, and NP formed (Table 3), the lowest NF was obtained in the control and -N omission, whereas the ND was lowest with -P, -Fe, -Mn, and -Zn omission, and the least NP values were found in the -P and -Zn omission, and control treatments (Table 3). In the case of the -K omission, there was no effect on the NP; however, there was a greater miscarriage, which was reflected in the lowest values of NPP. Low NPP values were also observed in the treatments omitting -P and -Zn, and control.

In the plants infected with nematodes, -P and -Cu omission displayed 35% and 40% of plants with necrotic nodes, respectively, whereas plants with -Fe and -Zn omission were like the complete treatment (Table 4), indicating low Fe and Zn interaction with *A. besseyi* for this type of deformation in the plant. Favoretto and Meyer (2019) reported that stem deformation is characterized by node thickening, which is one of the main symptoms of infection with *A. besseyi*. The nutrient content of SDW in the presence of nematodes was generally higher (Table 5), except in the treatments omitting -N, -S, and -Mn. In the presence of *A. besseyi*, compared to the complete treatment, the treatments

Table 4. Total number of nodes and percentage of necrotic nodes in response to the presence of *Aphelenchoides besseyi*.

Treatments	Nodes (total) (n)	Necrotic nodes (n)	Necrotic nodes (%)
Control	46b	16a	35a
Complete	67b	7b	10d
– N	50b	8b	16c
– P	48b	17a	35a
– K	63b	11b	17c
– S	84a	11b	13c
– B	74a	18a	24b
– Cu	50b	20a	40a
– Fe	89a	8b	9d
– Mn	86a	12b	14c
– Zn	76a	6b	8d
Mean	66.6	12.2	20.1
CV (%)	18.9	19.3	18.4

Note: *Means followed by the same lowercase letters in columns do not differ by the Scott-Knott test ($p \leq .05$).

Table 5. Nutrient content in shoot dry weight (SDW) of soybean at the reproductive growth stage R1-R2, due to fertilization treatments in the presence and absence of nematodes.

Nutrients	Treatments											
	Control	Complete Nutrition	-N	-P	-K	-S	-B	-Cu	-Fe	-Mn	-Zn	Média
(+) nematode												
N (g kg^{-1})	36.28a	39.92a	37.45a									37.88
P (g kg^{-1})	9.06a	10.56a		8.21a								9.28
K (g kg^{-1})	56.63a	49.09a			29.39a							45.04
S (g kg^{-1})	1.68a	1.60a				1.46a						1.58
B (mg kg^{-1})	5.44b	15.42a					6.29b					8.63
Cu (mg kg^{-1})	4.20b	12.41a						5.05b				7.22
Fe (mg kg^{-1})	61.22b	121.12a							62.58b			81.64
Mn (mg kg^{-1})	288.80b	383.86a								310.22b		327.63
Zn (mg kg^{-1})	57.97c	151.52a									70.63b	93.37
(-) nematode												
N (g kg^{-1})	29.00c	36.97b	46.33a									37.43
P (g kg^{-1})	4.60b	9.53a		3.22b								5.78
K (g kg^{-1})	18.43b	43.61a			20.67b							27.57
S (g kg^{-1})	1.12b	1.13b				1.53a						1.26
B (mg kg^{-1})	5.11b	14.33a					5.92b					8.45
Cu (mg kg^{-1})	1.13b	10.20a						1.22b				7.52
Fe (mg kg^{-1})	100.71b	104.81a							73.47c			93.00
Mn (mg kg^{-1})	261.45b	341.54a								368.00a		323.66
Zn (mg kg^{-1})	71.73b	131.91a									35.75c	79.80

Note: *Means followed by the same lowercase letters in columns do not differ by the Scott-Knott test ($p \leq .05$).

with -Cu, -Fe, -Mn, and -Zn omission showed significant differences (Table 5). In the infected plants that received complete nutrient treatment, except for B, the concentrations of the other nutrients studied were less than those found in plants without *A. besseyi*. In the absence of the pathogen, the nutrient content in all the omission treatments differed from that in the control (without nutrients) and complete nutrition treatment (Table 5). According to Almeida, Santos, and Martins (2011), the insufficient supply of nutrients can be owing to the damage caused by root and/or shoot nematodes. Thus, the mineral composition of plants parasitized by nematodes differs from that of the so-called healthy plants, whose physiological changes may not follow a rigid pattern, and in some cases, decrease or accumulation may occur, or the contents may remain unchanged without the effect of parasitism (Hussey 1985).

Conclusions

The presence of *A. besseyi* alters the growth characteristics of soybean plants, except for the treatment omitting -P, -S, -B, -Mn, and -Zn. The presence of nematodes led to reduced RL in plants with treatments omitting -K, -S, -B, -Cu, -Fe, -Mn, and -Zn and lesser RFW with -N, -P, -K, and -Cu omission. The total N assimilated by the plants did not differ significantly between treatments without the nematode, whereas its presence affected the control and complete nutrition treatments. The LAI was the lowest for inoculated plants treated without nutrition, complete nutrition, and -N, -P, -K, and -Cu omission. The pathogen influenced the agronomic characteristics of the inoculated plants where only nodes were examined and up to 40% of these were necrotic, while in the uninoculated plants, changes were observed in the number of nodes, flowers, small pods, and pods per plant. The nutrient concentrations were generally higher in the shoots in the presence of nematodes, except in the plants with -N, -S, and -Mn omitted treatments. The nutritional status of soybean does not influence infection by the nematode *A. besseyi*, but with nutrient application, soybean development is balanced, which in turn can reduce the severity of the disease.

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No potential conflict of interest was reported by the author(s).

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References

Almeida, E. J., J. M. Santos, and A. B. G. Martins. 2011. Influence of *meloidogyne enterolobii* parasitism on leaf nutritional status and phenology of 'paluma' guava plants. *Bragantia* 70 (4):876–81. doi:10.1590/S0006-87052011000400021.

Almeida, A. M. R., and C. D. S. Seixas. 2010. Soybean: Root and stem diseases and interrelationships with soil and crop management *[Soja: Doenças radiculares e de hastes e inter-relações como manejo do solo e da cultura]*. Londrina, Brazil: Embrapa Soja.

Alves, B. J. R., J. C. Franchini, A. D. Virgens Filho, J. O. A. Sena, A. Moreira, and E. Malavolta. 2002. Evaluation of macro and micronutrient availability to the upland rice cropped on a calcareous soil of Irecê region, Bahia. *Revista Universidade Rural Série Ciências Exatas E da Terra* 22 (1):15–25.

Bahry, C. A., M. Venske, E. Nardino, S. S. Fin, P. D. Zimmer, V. Q. Souza, and B. O. Caron. 2013. Efeito do nitrogênio suplementar sobre os componentes de rendimento da soja em condição de estresse hídrico. *Revista Agrarian* 6 (2):281–88. doi:10.1590/S0034-737X2014000200019.

Barreto, C. F., P. S. Silva, R. Navroski, J. A. Benati, G. Nava, and L. E. C. Antunes. 2017. Deficiência DE nutrientes com efeitos no desenvolvimento DE morangueiros. *Scientia Agraria* 18 (4):63–71. doi:10.5380/rsa.v18i4.53470.

Carlos, L., N. Venturin, R. L. G. Macedo, E. M. Higashikawa, M. B. Garcia, and E. S. Farias. 2014. Crescimento e nutrição mineral de mudas de pequi sob efeito da omissão de nutrientes. *Ciência Florestal* 24 (1):13–21. doi:10.5902/1980509813318.

Coolen, W. A., and C. J. D´Herde. 1972. *A method for the quantitative extraction of nematodes from plant tissue*. Ghent: State Agriculture Research Center.

Dias, W. P., G. L. Asmus, J. F. V. Silva, A. Garcia, and G. E. S. Carneiro. 2010. Nematodes [Nematoides]. In Soybean: Root and stem diseases and interrelationships with soil and crop management [Soja: Doenças radiculares e de hastes e inter-relações como manejo do solo e da cultura]. In ed. A. M. R. Almeida and C. D. S. Seixas, 173–206. Londrina, Brazil: Embrapa Soja.

Dias, J. P., W. P. Dias, R. B. Loreto L, L. A. Favoreto, C. Moraes, and A. Moreira. 2021. Effects of nutrient omission on the reproduction of meloidogyne javanica in soybean. *Journal of Plant Nutrition* 44 (14):2057–68. doi:10.1080/01904167.2021.1889594.

Embrapa (Empresa Brasileira de Pesquisa Agropecuária). 1997. Manual for methods of soil analysis *[Manual de Métodos de Análise de Solo]*. Rio de Janeiro, Brazil: Embrapa Solos.

Fageria, N. K., and A. Moreira. 2011. The role of mineral nutrition on root growth of crop plants. *Advances in Agronomy* 110:251–331.

Favoreto, L., J. M. Santos, S. A. Calzavara, and L. A. Lara. 2011. Phytosanitary study, multiplication and taxonomy of nematodes associated with seeds of forage grasses in Brazil. *Nematologia Brasileira* 35:20–35.

Favoretto, L., and M. C. Meyer. 2019. *The green stem nematode [O nematoide da haste verde]*. Londrina, Brazil: Embrapa Soja.

Fehr, W. R., C. E. Caviness, D. T. Burmood, and J. S. Pennington. 1971. Stage of development descriptions for soybeans, *Glycine max* (L.) Merrill. *Crop Science* 11 (6):929–31. doi:10.2135/cropsci1971.0011183X001100060051x.

Ferraz, S., L. G. Freitas, E. A. Lopes, J. C. O. Freitas, and J. O. D. Souza Júnior. 2012. Características morfofisiológicas de plantas clonais de *Passiflora alata* crescidas em diferentes doses de nitrogênio e níveis de sombreamento. *Revista Brasileira de Fruticultura* 34 (3):859–72. doi:10.1590/S0100-29452012000300028.

Hussey, R. S. 1985. Host-parasite relationships and associated physiological changes. In An advanced treatise on *meloidogyne*: Biology and control, ed. J. N. Sasser and C. C. Carter, 143–53. Raleigh, USA: North Carolina State University.

Kepenekci, I. 2013. Rice white tip nematode (*Aphelenchoides besseyi*) in rice growing areas of Turkey. *Nematropica* 43:181–89.

Maia, J. T. L., F. P. G. Bonfim, R. E. M. Guanabens, R. TRENTIN, H. E. P. Martinez, P. R. G. Pereira, and P. C. R. Fontes. 2014. Omissão de nutrientes em plantas de pinhão-manso cultivadas em solução nutritiva. *Ceres* 61 (5):723–31. doi:10.1590/0034-737X201461050016.

Malavolta, E. 2006. *Manual of mineral nutrition of plants [Manual de Nutrição Mineral de Plantas]*. São Paulo, Brazil: Agronômica Ceres.

Malavolta, E., G. C. Vitti, and S. A. Oliveira. 1997. *Evaluation of nutritional status of plants; principles and applications*, Avaliação do estado nutricional das plantas; princípios e aplicações. Piracicaba, Brazil: Potafos,

Marschner, P. 2012. *Marschner's mineral nutrition of higher plants*. London. UK: Academic Press.

Meyer, M. C., L. Favoreto, D. Klepker, and F. C. Marcelino-Guimarães. 2017. Soybean green stem and foliar retention syndrome caused by *Aphelenchoides besseyi*. *Tropical Plant Pathology* 42:403–09. doi:10.1007/s40858-017-0167-z.

Moreira, A., N. K. Fageria, A. Garcia, and Y. Garcia. 2011. Effect of liming on the nutritional conditions and yield of alfalfa grown in tropical conditions. *Journal of Plant Nutrition* 34 (8):1107–19. doi:10.1080/01904167.2011.558155.

Moreira, A., J. C. Franchini, L. A. C. Moraes, and E. Malavolta. 2000. Evaluation of nutrients in a calcareous vertisol. *Pesquisa Agropecuária Brasileira* 35 (10):2107–13. doi:10.1590/S0100-204X2000001000024.

Moreira, A., L. A. C. Moraes, T. R. Melo, R. Heinrichs, and L. G. Moretti. 2022. Management of copper for crop production. *Advances in Agronomy* 173:257–98.

Moreira, A., L. A. C. Moraes, and A. R. Reis. 2018. The molecular genetics of zinc uptake and utilization efficiency in crop plants. In *Plant micronutrient use efficiency: Molecular and genomic perspectives in crop plants*, ed. M. A. Hossain, T. Kamiya, D. J. Burritt, L. S. P. Tran, and T. Fujiwara, 87–108. London, UK: Elsevier/Academic Press.

Prado, R. M., C. F. Franco, and A. P. Puga. 2010. Macronutrient deficiencies in soybean cv. BRSMG 68 (Vencedora) cultivated in nutritive solution. *Comunicata Scientiae* 1 (2):114–19.

Zambolim, L., H. Costa, and X. R. F. 2001. *Integrated phytosanitary management: Protected cultivation, center pivot and no-till* [Manejo integrado fitossanidade: Cultivo protegido, pivô central e plantio direto]. In *Effect of mineral nutrition on plant diseases caused by soil pathogens [Efeito da nutrição mineral sobre doenças de plantas causadas por patógenos de solo]*, ed. L. Zambolim, 347–408. Viçosa, Brazil: UFV/DFP.