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ORIGINAL RESEARCH PAPER

# Entomopathogenic nematode distribution and edaphoclimatic conditions in the Cerrado of Minas Gerais, Brazil

**Vanessa Andaló[1] · Jéssica Mieko[1] · Fábio Janoni Carvalho[1] · Gleice Aparecida de Assis[1] · Lucas Silva de Faria[1] · Franscinely Aparecida de Assis[2] · Ronaldo Antônio dos Santos[1] · Francielle Rosa[1]**

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**Abstract**

Based on the high diversity of Brazilian fauna and flora, edaphoclimatic conditions in the Cerrado of Minas Gerais, and the situation of utilization of EPNs in Brazil, a survey was conducted in order to relate the presence of these organisms with the physical and chemical attributes of the soil, combined with precipitation. To this end, soil samples were collected in areas with diversified vegetation types in Monte Carmelo, MG, at Juliana Farm. The samples were obtained every 15 days for 6 months. From each spot, soil samples (about 500 g) were collected for soil moisture characterization, nematode isolation and determination of pH, organic matter, potassium (K), calcium (Ca), magnesium (Mg), phosphorus (P), potential acidity (H + Al) and precipitation data (mm). Three populations of entomopathogenic nematodes of the *Heterorhabditis amazonensis* species were isolated in Cerrado *stricto* sensu and Gallery forest areas. The occurrence *H. amazonensis* could not be considered restricted to specific soil condition, as organic matter, humidity, pH, Ca, K, Mg and H + Al, especially considering the organic matter and K values, which had variable levels between the places of collection. The *p* values of the positive soil samples were at a lower level than the mean of the Gallery forest and Cerrado, and at the same level as maize and pasture area. The soil moisture in the Cerrado area increased with the higher values of precipitation; however, in the Gallery forest area this association was not observed. Also, the nematodes were isolated when the temperature began to decrease.

**Keywords** *Heterorhabditis* · Soil analysis · Survey · Vegetation cover

## Introduction

The entomopathogenic nematodes have great potential as biological control agents for insect pests, supporting integrated pest management programs. The two main families of entomopathogenic nematodes are Steinernematidae and Heterorhabditidae, belonging to the order Rhabditida (Nematoda: Chromadorea) (Ferraz 1998). The development of studies with *Heterorhabditis* Poinar and *Steinernema* Travassos has enabled their use as a biological tool for pest control of various crops (Bellini and Dolinski 2012; Giometti et al. 2011).

New species of nematodes have been described in Brazil, such as *Heterorhabditis amazonensis* Andaló, Nguyen and Moino and *Steinernema brazilense* Nguyen, Ginarte and Leite (Andaló et al. 2006; Nguyen et al. 2010). The use of these species is closely related to the adaptation of the nematode to the environment and their targeting of specific insects (Andaló et al. 2014; Kaya and Gaugler 1993).

According to Leite et al. (2017) few nematodes are mass produced in Brazil, however studies with biodiversity, commercial production and formulation are increasing. Brida et al. (2017) obtained from agricultural soils with annual, fruit and forest crops in Brazil different species of nematodes, including *H. amazonensis*, *Metarhabditis rainai* (Carta and Osbrink), *Oscheios tipulae* (Lam and Webster) and *Steinernema rarum* (De Doucet). Dolinski et al. (2012) discussed the control of *Conotrachelus psidii* (Marshall) (Coleoptera: Curculionidae) in guava orchads, *Psidium guajava* L. (Myrtaceae), and observed a reduction on the number of damaged fruits, when applying the nematode *H. baujardi* Phan, Subbotin, Nguyen and Moens. However,

✉ Vanessa Andaló
vanessaandalo@ufu.br

[1] Universidade Federal de Uberlândia, Campus Monte Carmelo, LMG 746, Km 01, Monte Carmelo, MG CEP 38500-000, Brazil

[2] Universidade Federal de Lavras, Campus Universitário, Caixa Postal 3037, Lavras, MG CEP 37200-000, Brazil

observing the distribution of the isolated species in the world compared to the number of native Brazilian strains, highlights the importance of surveys in Brazil, considering the recognized diversity of fauna and flora existing in the different Brazilian ecosystems, including areas of native vegetation, and the propitious edaphoclimatic conditions in the Cerrado of Minas Gerais.

Entomopathogenic nematodes have the ability to locate and invade the body of a host insect through its natural openings and cuticles. Within the body of the insect, they release symbiotic bacteria (Steinernematidae: *Xenorhabdus* Thomas and Poinar, and Heterorhabditidae: *Photorhabdus* Boemare, Akhurst and Mourant, causing sepsis and death of the host in 48 h. Inside the body of the dead insect, the bacterium multiplies rapidly. The nematode then begins to feed on bacteria and reproducing inside the body of the dead insect. After two or three generations, when the food runs out, the infective juvenile (IJ) leaves the cadaver (Han and Ehlers 2000).

After the emergence of the IJs from the insect's cadaver, they search for new hosts. Once in the soil, these juveniles are subject to factors that may influence their survival and mobility, such as predators, ultraviolet radiation, extreme temperatures, soil moisture and texture, osmotic stress and pesticides (Brown and Gaugler 1997; Glazer 2002).

The main limiting factors for the ongoing maintenance of entomopathogenic nematodes populations are the rates of irradiation of ultraviolet light and availability of water in the soil. Other factors such as soil texture, temperature, the presence of agricultural crops and availability of hosts also influence their distribution in the soil. The presence of undergrowth vegetation or decaying vegetation cover favors soil protection and humidity maintenance, and this combination can provide an ecosystem favorable to the presence and persistence of these nematodes (Burman and Pye 1980; Kung et al. 1990; Woodring and Kaya 1988).

In accordance with the aforementioned aspects, the objective of this study was to survey the natural occurrence of entomopathogenic nematodes populations in an area with different vegetation coverings, in order to relate the presence of these organisms to the physical and chemical attributes of the soil, associated with precipitation data.

## Materials and methods

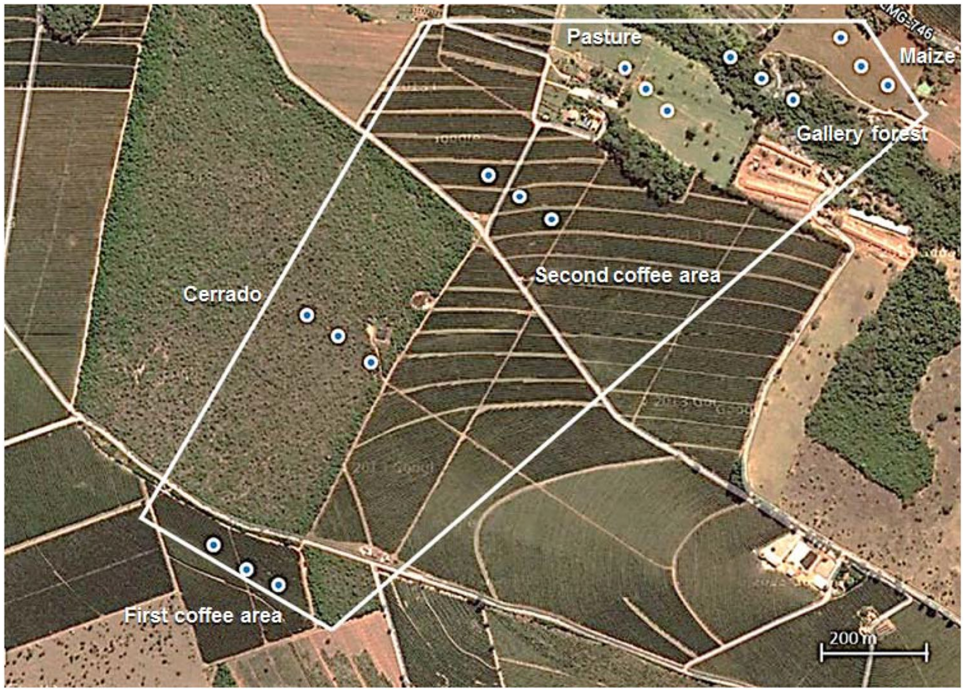
### Soil samples

Soil samples for nematode isolation were collected from areas of coffee (*Coffea arabica* L. (Rubiaceae)) and maize (*Zea mays* L. (Poaceae)) crops, pasture (*Brachiaria decumbens* (Hochst. ex A. Rich.) Stapf (Poaceae)), from vegetation of Cerrado *stricto* sensu (*Anacardium occidentale* L. (Anacardiaceae), *Jacaranda* spp. Juss. (Bignoniaceae), *Annona* spp. Mill. (Annonaceae), *Qualea* spp. Aubl. (Vochysiaceae), *Caryocar brasiliense* Cambess. (Caryocaraceae), *Tabebuia* spp. Gomes ex DC. (Bignoniaceae)) and Gallery forest (*Tibouchina* Aubl. (Melastomataceae), *Xylopia* spp. L. (Annonaceae), *Dendropanax cuneatus* (DC.) Decne. and Planch. (Araliaceae), *Mauritia flexuosa* Mart. (Arecaceae), *Plenckia populnea* Reissek (Celastraceae), *Calophyllum brasiliense* Cambess. (Calophyllaceae), *Tabebuia* spp., *Clusia nemorosa* G. Mey. (Clusiaceae), *Kielmeyera coriacea* Mart. & Zucc. (Clusiaceae)) (Medeiros 2011) in the county of Monte Carmelo, MG, Brazil, at Juliana farm, geographic coordinates 18°42′5.83″S, 47°33′0.27″W (Fig. 1), altitude 880 m, located in the domain of Plateau and Tablelands of the Paraná Sedimentary Basin, in the Southwest portion of the Brazilian Cerrado biome.

According to the classification of Köppen, the climate of the region is of the Cwa type, with an average annual temperature of 22 °C and annual average rainfall of 1500 mm, with rains concentrating in a 6 month period, mainly in the summer (Oliveira 2010). The soil type is predominantly red clay latosol.

The area cultivated with Arabica coffee is irrigated by fertigation, with planting spaced at 3.8 × 0.7 m. Acaricide propargite, inseticide/fungicide ciproconazole + thiamethoxam, insecticide clorantraniliprole + lambda-cyhalothrin, insecticide/acaricide abamectin, fungicides azoxystrobin and thiophanate-methyl and herbicides glyphosate and chlorimuron-ethyl are routinely used in the coffee areas. The herbicides glyphosate, atrazine, nicosulfuron and the insecticide chlorpyrifos were applied in the area cultivated with maize. No pesticides were applied in the pasture area. Chemical fertilizers added to the soil in the agricultural areas were $CO(NH_2)_2$ (45% N), $NH_4H_2PO_4$ (12% N, 60% $P_2O_5$), KCl (60% $K_2O$).

In order to collect soil, points were marked from the south to the north of the terrain in six sequential areas: a first coffee area (7.1 ha), Cerrado (22 ha), a second coffee area (9.7 ha), Gallery forest (12.1 ha), pasture (8.4 ha) and maize (8.7 ha). The total area sampled comprises 68 ha. The two coffee areas were considered separately for soil samples due to the distance between them in the property. For each area, three samples were collected per day, totaling 18 soil samples per day of collection (six areas × three samples) (Fig. 1). Within each collection area the points were distributed longitudinally every 50 m. Samples were collected every 15 days, over 6 months, totaling 12 evaluations for each area sampled (two evaluations per month), during the spring, summer and autumn, beginning on November 04, 2014 and ending on April 17, 2015. The three samples were collected from different points in each survey. The total sampling points were 216 (18 samples per day × 12 days of evaluation).




• Distribution of sampling points.

**Fig. 1** Location of soil sample collection. Juliana farm, Monte Carmelo—MG, 18°42′5.83″S, 47°33′0.27″W. Demarcated area corresponds to the sampled area of 68 ha. Juliana farm, Monte Carmelo, Minas Gerais, Brazil. Geographic coordinates 18°42′5.83″S, 47°33′0.27″W. *Source* Google Earth (2017)

Each soil sample contained about 500 g and was collected with a shovel from a depth between 5 and 15 cm. The samples were divided for isolation of entomopathogenic nematodes and physical and chemical characterization. Each sample was packed in a plastic bag and kept in a Styrofoam box at about 20 °C for 2 h until being processed in the Entomology Laboratory of the Federal University of Uberlândia. Samples were tagged with date and place of collection, crop or associated vegetation and the geographical coordinates of the site.

## Entomopathogenic nematode isolation

In the laboratory, to isolate entomopathogenic nematodes, the soil samples were processed through the insect-bait isolation technique (Stock 1998). About 200 g of soil was placed in a plastic container (14 × 14 × 7 cm) and moistened with 15% w/v of sterilized distilled water. Ten last-instar larvae of *Tenebrio molitor* L. (Coleoptera: Tenebrionidae) were placed on the moistened soil and the container was closed and inverted so that the larvae remained at the bottom. *Tenebrio molitor* is considered a sentinel host to isolate entomopathogenic nematodes and an insect with high mortality response (Caroli et al. 1996; Yan et al. 2016). The containers were kept at 24 ± 1 °C, and the larval mortality was evaluated after 5 days, verifying the characteristic symptomatology caused by nematode infection.

## Multiplication of the isolated material

The dead larvae with symptoms of infection were placed in a dry chamber (9 cm diameter Petri dish with filter paper) for 5 days, after which the larvae were removed and placed in White traps (White 1927) to collect IJs. To purify and confirm the parasitism, the Koch postulates were applied by multiplications in *T. molitor*. The traps were kept at 24 ± 1 °C. After Koch's postulates were fulfilled, a methodology of Molina and López (2001) was applied to multiply these isolated native nematodes, and the next generations were obtained for identification. *Tenebrio molitor* larvae were reared according to the methodology of Potrich et al. (2007).

## Physical and chemical characteristics of soil and precipitation data

Soil samples were used to perform physical and chemical analyses. To determine moisture, a soil sample of about 60 g was added to an aluminum can of known weight and placed in a drying oven at 105 °C for 24 h; after that, the dry weight was obtained.

The samples were submitted to chemical analysis to determine pH, organic matter, potassium (K), calcium (Ca), magnesium (Mg), phosphorus (P) and potential acidity (H + Al), according to the methods described by Embrapa (1997). The precipitation data (mm) were obtained from the nearest meteorological station, located in Monte Carmelo, MG, Brazil, 8 km away from the experimental area, in the periods of sampling, in order to verify the possible influence of these factors on the occurrence of nematodes.

### Statistical analysis

The data obtained regarding the soil characteristics were submitted to analysis of variance and the Scott–Knott test at 5% probability for comparison between means, using the statistical software SISVAR® version 5.0 (Ferreira 2011).

## Results

Three populations of entomopathogenic nematodes were found through morphological and molecular techniques (Andaló et al. 2006) and identified as *H. amazonensis*. Two populations were found in the Cerrado area and one in the Gallery forest. Both areas are characterized by the presence of high density vegetation cover, due to the characteristics associated with these biomes. Among the three populations of nematodes isolated, two of them were obtained on February 17, 2015 (one in the Cerrado area, geographic coordinates 18°41′48″S, 47°33′28″W and the other in the Gallery forest area, 18°41′52″S, 47°33′70″W); and the other population was obtained on March 03, 2015 (in the Cerrado area, 18°41′49″S, 47°33′27″W).

The data from physical and chemical characteristics of the soil in the survey areas were considered distinct. Regarding soil moisture, a difference between the coffee areas and the others was observed (Table 1). This could occur because the coffee areas were drip irrigated, increasing humidity in comparison to the other areas.

The coffee areas also presented the highest values of pH and the lower values of potential acidity (H + Al) (Table 1). This difference could be attributed to the limestone application for pH calibration in interspersed years in coffee areas, while in other areas this operation was not performed.

The P content in Cerrado, Gallery forest and coffee areas soils was different significantly from that in Pasture and Maize soils. There was no significant difference in K and Mg levels in the sampled areas (Table 1).

Considering the areas with the positive sample for nematodes, it was obtained that in the Gallery forest area the humidity in February (10.15 g) was higher than the data found in the Cerrado at the same date (7.62 g). The positive soil sample from Gallery forest was with the highest content of organic matter (Table 2), which could promote the maintenance of soil moisture. While in March the same Cerrado area had a value (10.84 g) similar to the Gallery forest in February (Table 2), which could occur because of the increase of precipitation on these periods (Fig. 2), which were 169.0 mm in February 17, 2015 and 202.6 mm in March 03, 2015.

Regarding pH, the obtained values in the samples were between 5.9 and 6.9, from which it is difficult to conclude about the influence of this factor, since no inference at larger pH scales was possible (Table 2). Regarding soil acidity, it was observed that entomopathogenic nematodes were found in areas with a variation of H + Al (cmolc $dm^{-3}$) in the soil (Table 2). So the acidity of the soil was not a limiting factor for the occurrence of these organisms.

Considering soil nutrients, it was observed that entomopathogenic nematodes were present in regions with a diverse content of Ca (cmolc $dm^{-3}$), as well as for K, the nematodes were obtained in areas with variable content of this nutrient. The P content in the positive nematode samples was considered low, at the same level as pasture and maize areas. There was no difference between Mg contents in soils with isolated nematodes (Table 2).

During the collection months, the occurrence of rain was irregular varying among the periods of collection.

**Table 1** Physical and chemical characteristics of the soil in the areas of the entomopathogenic nematode survey

| | Organic matter (dag $kg^{-1}$) | Humidity (g) | pH (in water) | Ca (cmolc $dm^{-3}$) | P $meh^{-1}$ (mg $dm^{-3}$) | K (cmolc $dm^{-3}$) | Mg (cmolc $dm^{-3}$) | H + Al (cmolc $dm^{-3}$) |
|---|---|---|---|---|---|---|---|---|
| Gallery forest | 3.81 ± 0.106 a | 8.37 ± 0.149 b | 6.35 ± 0.041 b | 4.08 ± 0.147 a | 56.36 ± 2.164 a | 0.390 ± 0.017a | 1.60 ± 0.066 a | 2.54 ± 0.122 a |
| Cerrado | 3.75 ± 0.190 a | 8.67 ± 0.109 b | 6.19 ± 0.067 b | 3.63 ± 0.212 a | 68.43 ± 7.203 a | 0.510 ± 0.020 a | 1.27 ± 0.039 a | 3.21 ± 0.199 a |
| Pasture | 3.18 ± 0.084 b | 8.14 ± 0.144 b | 6.20 ± 0.044 b | 2.71 ± 0.073 b | 12.22 ± 0.812 b | 0.302 ± 0.016 a | 1.49 ± 0.045 a | 3.36 ± 0.161 a |
| Maize | 2.94 ± 0.108 b | 8.47 ± 0.270 b | 6.06 ± 0.043 b | 2.68 ± 0.101 b | 29.81 ± 3.568 b | 0.401 ± 0.009 a | 1.02 ± 0.067 a | 2.99 ± 0.106 a |
| First Coffee Area | 2.69 ± 0.054 b | 10.84 ± 0.182 a | 6.81 ± 0.052 a | 4.25 ± 0.072 a | 58.75 ± 2.682 a | 0.307 ± 0.011 a | 1.37 ± 0.019 a | 1.56 ± 0.042 b |
| Second Coffee Area | 2.50 ± 0.041 b | 9.98 ± 0.145 a | 6.86 ± 0.043 a | 3.87 ± 0.100 a | 60.83 ± 4.078 a | 0.341 ± 0.015 a | 1.53 ± 0.046 a | 1.83 ± 0.067 b |

Juliana Farm, Monte Carmelo, MG, Brazil. Means (± SE) followed by the same letter in the column do not differ from each other by the Scott–Knott test at 5% significance

**Table 2** Physical and chemical characteristics of the soil in the areas of positive soil samples for entomopathogenic nematodes

| Field | Organic matter (dag kg$^{-1}$) | Humidity (g) | pH (in water) | Ca (cmolc dm$^{-3}$) | P meh$^{-1}$ (mg dm$^{-3}$) | K (cmolc dm$^{-3}$) | Mg (cmolc dm$^{-3}$) | H + Al (cmolc dm$^{-3}$) |
|---|---|---|---|---|---|---|---|---|
| Cerrado (February 17, 2015) | 2.70 | 7.62 | 6.00 | 3.10 | 26.30 | 0.24 | 1.40 | 2.70 |
| Gallery forest (February 17, 2015) | 5.40 | 10.15 | 6.90 | 5.80 | 19.60 | 0.80 | 1.80 | 1.30 |
| Cerrado (March 3, 2015) | 2.30 | 9.53 | 5.90 | 1.70 | 19.30 | 0.28 | 1.00 | 4.30 |

Juliana Farm, Monte Carmelo, MG, Brazil

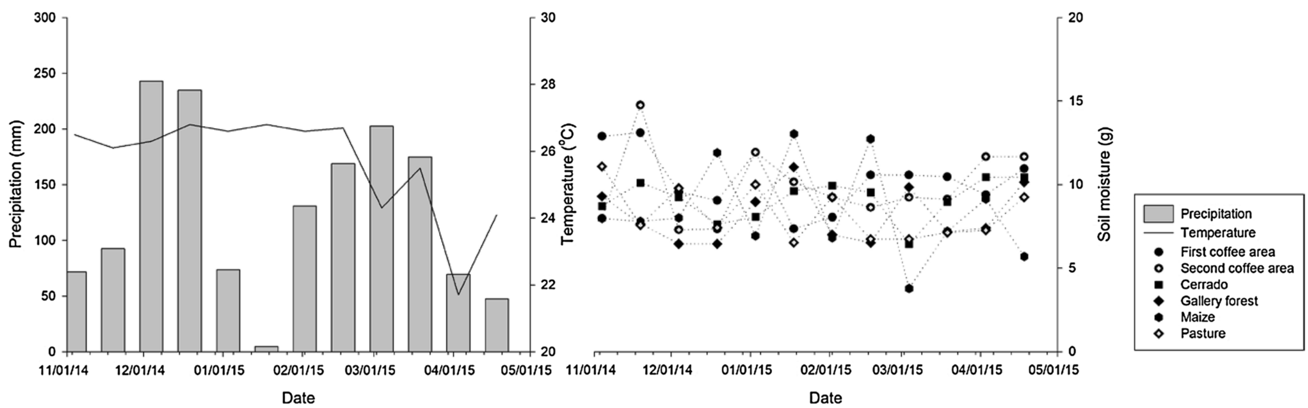



**Fig. 2** Precipitation (mm), temperature (°C) and soil humidity (g) in the period of the collections. Nematode isolation occurred at February 17, 2015 and March 3, 2015 in Cerrado areas; and at March 3, 2015 in the Gallery forest area

Nematodes were collected in periods of intermediate precipitation and temperature rates, about 160 mm per month and average temperature of 26 °C. In February and March the means of precipitation and temperature were 149.9 and 188.7 mm and 26.6 and 24.9 °C, respectively. At the dates that positive samples were obtained, February 17, 2015 and March 03, 2015, these values were 169.0 and 202.6 mm and 26.7 and 24.3 °C, respectively. Soil moisture was highly variable according to the area and period of evaluation (Fig. 2).

## Discussion

The survey performed in the Monte Carmelo region, Brazil, resulted in three nematode populations, all of them were identified as *H. amazonensis*. The populations were isolated from areas with a higher vegetation cover compared to the other areas, as pasture and maize. However, Griffin et al. (2000) verified that *H. indica* Poinar, Karunakar and David does not restrict its distribution as a function of the vegetation cover. The same could occur with *H. amazonensis*, since this species was previously isolated in the Amazonian forest (Andaló et al. 2006) and in sorghum and garlic cultivated areas (Andaló et al. 2009).

Hazir et al. (2003) suggested that entomopathogenic nematodes could be more prevalent in crop areas rather than natural environment areas, depending on population peaks and availability of different insect populations. However, the occurrence of *H. amazonensis* in the sampled areas of natural vegetation could also be associated with the presence of insects in the soil, since insects in areas of Cerrado and Gallery forest are diverse (Dias 1996; Myers et al. 2000), but more distributed over time, depending on the balance of the environment.

Yet another aspect that should be highlighted is the fact that the crop areas with coffee and maize received a lot of agricultural inputs, as insecticides and herbicides, which may interfere negatively with the survival of nematodes. Although this aspect was not studied in the present work, Negrisoli et al. (2010) observed that the product Verdadero® reduced the infectivity of *Heterorhabditis bacteriophora* Poinar, but not of *Steinernema carpocapsae* (Weiser), which demonstrated that the influence of pesticides on nematodes could vary with the nematode species. However, Sabino et al. (2014) verified that the product Ampligo® did not cause damage to *H. amazonensis*. So, it is possible to infer that the diversity of pesticides applied in the studied area could interfere in the presence of entomopathogenic nematodes.

Besides the presence of the host insect, several other factors, biotic and abiotic, as well as biological characteristics of the species can influence the presence and survival of entomopathogenic nematodes in the soil. Campos-Herrera et al. (2016) stated that the presence of entomopathogenic nematodes obtained in surveys performed in natural areas is considered high in relation to surveys conducted in crop areas, but conclude that the comparisons are imprecise since the sampling methods used are very diverse. Techniques, such as qPCR, apply specific molecular probes to recognize entomopathogenic nematodes that inhabit the soil, but surveys are frequently performed according to the insect-bait technique, using *Galleria mellonella* L. (Lepidoptera: Pyralidae) or *T. molitor* as sentinel hosts, which makes it difficult to compare the results since the method could influence the diversity and abundance of entomopathogenic nematodes sampled.

The nematodes species were isolated from soils with variable organic matter contents, highlighting the higher content for the Gallery forest area (5.4 dag $kg^{-1}$) (Table 2). Although Campos-Herrera et al. (2013) observed that the characteristics that regulate soil moisture, such as organic matter, are potential regulators of the spatial distribution of entomopathogenic nematodes. Alumai et al. (2006) and Campos-Herrera et al. (2016) verified that entomopathogenic nematodes are associated with moderate organic matter, P and Mg and highlight the correlation between soil properties, such as water potential, soil pH and fertility, as determinants for nematode distribution.

One nematode population was isolated from a soil in conditions with high moisture and low organic matter content in Cerrado (Table 2). Thus, the nematode adaptation to soil moisture helps in the management strategies for pest control (Duncan et al. 2013; Campos-Herrera et al. 2014). The studied coffee areas are irrigated throughout the year, which facilitates the release of EPNs through this process, allowing the higher survival of these organisms in the soil, since the humidity remains high; in addition, it is a process already conducted in the field, reducing application costs. Shapiro-Ilan et al. (2015) and Arrington et al. (2016) discussed that the use of irrigation systems to apply EPNs that can enhance their efficacy to control insect-pests.

According to Alumai et al. (2006), entomopathogenic nematodes are usually found in areas with low pH, as well as in areas with high soil fertility and high sand content, and the interaction between these factors and the area management influences the occurrence and persistence of nematodes in the area. Campos-Herrera et al. (2013, 2014) verified that soil pH influenced the occurrence of entomopathogenic nematodes, and increasing soil pH caused a reduction in the abundance of steinernematid species in citrus orchards, but did not influence *H. indica*. Kung et al. (1990) demonstrated that soil pH and $O_2$ availability influenced IJ survival; soils with pH less than 10 and well aerated, without excessive irrigation or frequent flooding, enable the presence of these organisms, which demonstrates that there is a close correlation between the soil characteristics and nematode distribution.

Calcium is an important macronutrient of the soil that acts by reducing soil acidity and decreasing the toxicity of aluminum (Al), copper (Cu) and manganese (Mn) (Nascimento and Lapido-Loureiro 2009). So an occurrence of entomopathogenic nematodes can be associated not only with the presence of these macronutrients, but also the correlation with other soil nutrients.

Potassium is an ion that has a positive charge, which leads to competition with Ca and Mg in the soil binding sites, so the Ca, Mg and K ratio must be balanced (Nascimento and Lapido-Loureiro 2009). Thus, the presence of K in the soil could influence the occurrence of entomopathogenic nematodes, not directly, but because of the strong correlation that it presents with other nutrients.

In studies with free-living nematodes, Hoy et al. (2008) affirmed that P is the abiotic factor with the highest correlation with the presence of these organisms. In addition, they verified that other soil attributes are correlated with each other and contribute in a similar way to the presence and abundance of nematodes in the soil. In the present study, all three nematodes were isolated from lower P condition soils in the higher P condition areas, Gallery forest and Cerrado (Tables 1, 2). Accordingly, P condition might be an abiotic factor in relation to the occurrence of *H. amazonensis* in the survey area.

In a survey of entomopathogenic nematodes, Alumai et al. (2006) obtained the species *H. bacteriophora*, *S. carpocapsae* and *Steinernema glaseri* (Ssteiner) Wouts, Mracek, Gerdin and Bedding and verified that the presence of these organisms was correlated with the content of P, organic matter and Mg, but there was no correlation with the pH, Ca and K. The authors concluded that the occurrence of entomopathogenic nematodes is more likely in areas with lower management, which receive fewer agricultural inputs and that have moderate levels of organic matter, P and Mg. These parameters are consistent with the areas where the populations of *H. amazonensis* were found. In addition, in these places the soil is not disturbed and does not receive agricultural inputs, which could have favored the presence of entomopathogenic nematodes.

It was observed that the precipitation is a parameter that influences the presence of nematodes in the soil, since they were isolated in periods of intermediate rainfall. Entomopathogenic nematodes need humidity to remain alive in the soil. The presence of these organisms in these periods may have occurred due to the fact that isolated populations were obtained from areas with relative vegetation cover, Cerrado and Gallery forest, which may have contributed to the maintenance of soil moisture. Risser et al. (2016) observed higher diversity and abundance of entomopathogenic nematodes in areas with higher precipitation levels, indicating the positive association of the presence of entomopathogenic nematodes and rainfall. Kanga et al. (2012) also isolated entomopathogenic nematodes in areas with high levels of precipitation, concluding that annual rainfall is a parameter associated with the presence of entomopathogenic nematodes; and concluded that the occurrence of entomopathogenic nematodes is related to a combination of several parameters, such as soil characteristics, moisture and climatic conditions.

Parameters such as precipitation and several soil properties could influence the composition of entomopathogenic nematode communities. Therefore, knowledge of the characteristics that favor each species allows for the manipulation of the environment in such a way that agricultural practices could be used to increase the persistence and the activity of these populations in the field.

Thus, further tests should be performed at different months of the year in order to obtain more positive samples, including the possible isolation of new species. In addition, analysis with other factors, such as soil organisms and pesticides, can contribute to elucidate unanswered questions, making the relationship between entomopathogenic nematodes and biotic and abiotic factors clearer.

## Conclusion

Natural populations of *H. amazonensis* were obtained in areas of Cerrado and Gallery forest in the region of Monte Carmelo, MG, Brazil. Among the evaluated attributes, the ones that influence the occurrence of nematodes were P, humidity, organic matter and precipitation.

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