Article

Effectiveness of commercial biological and chemical extracts for the control of nematodes in coffee tree in Chiapas

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Abstract

Phytoparasitic nematodes constitute one of the main pathogens that affect the cultivation of the coffee tree *Coffea arabica* L., causing reductions in yield between 15 and 60%. The objective of this research was to evaluate the effectiveness of the biological extracts of the nematicide Nemaxxion XT Plus, for the control of the nematodes *Meloidogyne incognita* and *Pratylenchus* sp., associated with the cultivation of coffee. Five treatments with five replications were considered under a randomized complete block design: Nemaxxion XT Plus (2 L ha^{-1}, 4 L ha^{-1}, 6 L ha^{-1}), Nemacur® 400 CE and control. Nematode populations were counted in 100 g of soil before applying the treatments and 120 days after the experiment was installed. For counting, the nematodes were extracted by the Baerman funnel method. For *M. incognita*, the number of second instar juveniles, adult males and females, was counted and for *Pratylenchus* sp., the filiform adult males and females were counted. The evaluation demonstrated the effectiveness of the treatments for the control of nematode populations, these treatments presented a considerable reduction of the populations, with an average survival of 1.2-3.4 nematodes of *M. incognita* and 2.2-4.4 nematodes of *Pratylenchus* sp., of these, Nemaxxion XT Plus at 6 L ha^{-1} presented the greatest population reduction and efficiency over the two populations, with 99.08 and 92.87% respectively, presenting itself as an alternative for a more sustainable agriculture.

Keywords: *Meloidogyne incognita*, *Pratylenchus* sp., injury nematode, root knot nematode.

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Introduction

The coffee or coffee tree, *Coffea* spp., is a plant belonging to the Rubiaceae Family and is native to Ethiopia. It was first cultivated in the province of Kaffa in Ethiopia in the middle of the 12th century, from where it spread through trade to the Middle East and from the 15th century it was introduced to Europe, where its consumption became widespread for the next 200 years (Perez and Díaz, 2000).

At present, more than one hundred species of *Coffea* are known, of which *Coffea arabica* L. and *Coffea canephora* L., are the two most important cultivated species, the first with a contribution of 58% of world production and the second with 42% (ICO, 2017). For 2014, the main green coffee producers in the world were Brazil with 2 804 070 t, Vietnam with 1 406 469 t, Colombia with 728 400 t, Indonesia with 643 900 t and Ethiopia with 419 980 t (FAO, 2017).

In Mexico, *Coffea arabica* L. occupies more than 97% of the coffee growing area and the robusta coffee tree *Coffea canephora* L. (Var. Robusta) is located to a lesser extent in the rest of the area, the latter aimed almost exclusively at the instant coffee sector. Currently there are about 120 varieties of Arabica coffee; highlighting the varieties Creole (Typica), Bourbon, Catimores, Caturra (red and yellow), Colombia, Costa Rica, Garnica, Maragogipe, Mundo novo, Oro azteca, Pacamara and Pluma hidalgo (AMECAFÉ, 2016).

In 2015, in 16 states of the Mexican Republic 1 026 251 98 t of coffee were produced, in an area of 734 291 03 ha, distributed in the mountain areas that cross the country, with a production value of $5 340 761 16. In this sense, the main producers with 64.2% of national production are the states of Chiapas and Veracruz with 383 059 62 (37.3%) and 276 054 71 (26.9%) tons respectively (SAGARPA-SIAP, 2017).

The presence of pests is of great importance in the cultivation of coffee, mainly due to their high incidence or the level of damage they cause. Phytoparasitic nematodes constitute one of the main limiting pathogens that affect coffee cultivation, particularly the *Coffea arabica* species that is susceptible to nematodes; capable of causing reductions in yield ranging between 15-60% (Campos and Villain, 2005; ANACAFÉ, 2017).

Numerous species of nematodes have been reported in association with this crop; however, the species of the genera *Pratylenchus Filipjev* (Tylenchida: Pratylenchidae) and *Meloidogyne Goeldi* (Tylenchida: Heteroderidae) are the most widely distributed and the most economically important, with the greatest losses in cultivation; the former with yield losses of 29-78% and the latter, yield losses of between 10-15% or up to 100% under certain conditions (Inomoto *et al*., 1998; Oliveira *et al*., 1999; Campos and Villain, 2005; Carneiro and Cofcewicz, 2008; Villain, 2008; Souza and Bressan-Smith, 2008).

The phytosanitary problem with nematodes is increasing, and the control alternatives are few, mainly systematic applications of chemical compounds are used, due to their effective effect on the different species of phytoparasitic nematodes (Perry, 1998; Baños *et al*., 2010). However, their use has been restricted by the harmful effects on the environment and they can become very toxic to the health of producers and consumers (Fe, 2002; Baños *et al*., 2010).

On the other hand, the cost, residuality and phytotoxicity of nematicides for cultivation limit their application, so it is essential to look for alternatives to combat phytopathogenic nematodes, which considerably affect production (Hernández *et al*., 2015).

Currently agriculture demands the reduction of chemical products and an increase in products that are friendly to the environment and human health; for this reason, biorational control; based on biological extracts and biological control organisms, it is presented as a very promising alternative, which allows the development of a more profitable agriculture that does not pollute the environment; This is why it is proposed to evaluate the biological effectiveness of Nemaxxion XT Plus, based on plant extracts and organic components for the control of the nematodes *Meloidogyne incognita* and *Pratylenchus* sp., associated with the cultivation of coffee in Motozintla, Chiapas.

Materials and methods

Location of the experiment

The field research was developed in the commercial production farm called Guadalupe Zaju, with the cultivation of coffee Var. Catuai in the fruit development stage, located in the producing region called 'El Soconusco', belonging to the municipality of Motozintla, State of Chiapas, with a silty crumb soil and an elevation of 1 000 meters above sea level, with a minimum average temperature of 22 ºC and a maximum of 29 ºC and an average monthly rainfall of 92 mm.

The research corresponding to the laboratory was carried out in the Nematology Laboratory of the Parasitology Department of the Autonomous Agrarian University Antonio Narro (UAAAN), in Saltillo, Coahuila, Mexico.

Initial sampling and identification of nematodes

In order to recognize the population of nematodes associated with the crop before the application of treatments, an initial sampling was carried out in two coffee trees per experimental unit, in the north cardinal point a sub sample of soil of 1 kg was taken at a depth of 40 cm in each tree, the two sub samples were mixed and only a representative sample of 1 kg of soil was taken, the 25 composite samples were transferred to the Nematology Laboratory; of each composite sample, 100 g were processed to obtain the nematodes present, using the Baerman funnel method (Cepeda, 1995).

The nematodes *Meloidogyne* sp. and *Pratylenchus* sp., were identified at the genus level based on the taxonomic keys of Cepeda (2016). Perineal cuts were made in adult females of the genus *Meloidogyne*, following the Taylor and Netscher (1974) technique, and the taxonomic keys of Eisenback *et al*. (1981), were used to corroborate the species, coinciding with the *Meloidogyne incognita* species.

Application of treatments

Directly in the coffee trees, channels (ditches) were opened in a circular way in the drip zone of the fruit, where the treatments to be evaluated were placed directly in contact with the root system, using a manual sprinkler backpack and later the channel was covered with the same soil.

A completely randomized block design (distance of 2 m between blocks) was established with five treatments and five repetitions and each repetition with an experimental unit represented by two two-year-old coffee trees, distributed at a planting distance of 1 m (Table 1).

Table 1. Treatments to evaluate the effectiveness of control of nematodes associated with the cultivation of coffee in Motozintla, Chiapas.

Treatments	Active ingredient (s)	Dose (L ha^{-1})
1	Biological extracts[1]	2
2	Biological extracts[1]	4
3	Biological extracts[1]	6
4	Fenamiphos[2]	3
5	Water[3]	0

[1]= products provided by GreenCorp Biorganiks de Mexico, SA de CV. Nemaxxion XT Plus= mixture of plant extracts of cempasúchil 2% (2 g ml^{-1}) *Tagetes erecta* L. (Asteraceae), Governor 5% (5 g ml^{-1}) *Larrea tridentata* L. (Sesse and Moc. Ex DC.) Coville (Zygophyllaceae), walnut shell 5% (5 g ml^{-1}) *Carya illinoinensis* (Wangenh.) K. Koch (Juglandaceae), essential oils of vegetable origin 10% (10 g ml^{-1}) chitosan 2% (2 g ml^{-1}), organic matter 5% (5 g ml^{-1}), leonardite fulvic acids 30% (30 g ml^{-1}) and emulsifiers and diluents 41% (41 g m^{-1}); [2]= Nemacur® 400 CE, systemic organophosphate, chemical control (equivalent to 35 g de ai. L^{-1}); [3]= absolute control.

During the development of the investigation, the nematicide Nemaxxion XT Plus was applied three times from the start of the experiment at intervals of 14 days between applications and the nematicide Nemacur 400 CE was applied only once.

The agronomic management of the crop during the development of the experiment was carried out based on the typical practices of the region, particularly cleaning activities and management of foliar diseases; mainly the coffee rust caused by the fungus *Hemileia vastatrix* (Berkeley & Broome) (Basidiomycota: Pucciniales), using copper oxychloride (cupric fungicides).

Final sampling

120 days after the last application of the treatments, in each of the experimental units, in the dripping area of the fruit tree, a final sampling was carried out in the same way as the initial sampling and the samples were sent to the Nematology laboratory, where the nematodes were extracted using the Baerman funnel method (Cepeda, 1995), identified and counted to obtain the final population.

With the results, the population increase index was related by the formula developed by Seinhorst (1970): I= Pf/Pi. Where: I= is the rate of population increase; Pf= is the final population and Pi= is the initial population.

In addition, the efficiency of the treatments was determined by the formula proposed by Henderson-Tilton (1955): E= (1-Pfa/Pia∗Pib/Pfb)∗100. Where: E= is the efficiency of the treatments, Pia= is the initial treatment population; Pfa= is the final treatment population; Pib= is the initial population of the control treatment and Pfb= is the final population of the control treatment.

The evaluation of the nematicidal activity (population reduction) was obtained through the difference between the initial population and the final population and was analyzed by means of an Anova with Tukey's separation of means ($p < 0.05$), using the statistical software SAS 9.0 (SAS, Institute, 2002).

Results and discussion

Table 2 shows the population at the beginning of the investigation and the final population found 120 days after the application of the treatments for the control of *Meloidogyne* spp. and *Pratylenchus* sp., obtained from the soil of the coffee plantation.

Table 2. Mean of the initial population and final population of *Meloidogyne incognita* and *Pratylenchus* sp., in each treatment, obtained from coffee soil.

Treatments	*Meloidogyne incognita*		*Pratylenchus* sp.	
	Initial	Final	Initial	Final
1	129.2	3.4	35.2	4.4
2	159	2.6	34.6	4.2
3	129.8	1.2	32.8	2.2
4	166.2	2.8	29.6	3.6
5	140.2	121.4	44.2	57.2

A significant reduction in the nematode population is observed as a result of the application of control treatments; while the control treatment presented an increase in the population in *Pratylenchus*; however, in *M. incognita*, the control presented a reduction of 12.84%, with respect to the initial population.

Table 3 shows the reduction of the population of *M. incognita* and *Pratylenchus* sp., in which it is observed that the treatments based on plant extracts and organic components, present a significant control ($p < 0.05$) over the different nematodes of the crop of coffee.

Table 3. Reduction of the populations of *Meloidogyne incognita* and *Pratylenchus* sp., after the application of treatments.

Treatments	Reduction of the nematode population (%)			
	Meloidogyne incognita		*Pratylenchus* sp.	
1	97.31	a	87.61	a
2	98.38	a	87.47	a
3	99.08	a	92.87	a
4	98.28	a	87.83	a
5	12.84	b	0	b

Population reduction in the same column with the same letter, they are not significantly different (Tukey <0.05).

Between the test treatments (1-4) for the control of *M. incognita*., no significant differences were found, with a control of 97.31 to 99.08% of the populations, while the control presented a significantly lower reduction, with a survival of 87.16% (gl= 8, 24; F= 334.12; $p < 0.0001$; R^2= 0.99, CV= 4.07) (Table 3).

The plant extracts of Nemaxxion XT Plus did not allow the population density of *M. incognita* to increase, reducing in a range of 0.009 to 0.026 times the initial population, mainly in treatment 3 (Nemaxxion XT Plus 6 L ha^{-1}), the which resulted with greater efficiency with 98.93% (Table 4).

Table 4. Index of population increase of *Meloidogyne incognita* and *Pratylenchus* sp., and efficiency of the treatments evaluated for the control of nematodes.

Treatments	*Meloidogyne incognita*		*Pratylenchus* sp.	
	Increase rate	Efficiency (%)	Increase rate	Efficiency (%)
1	0.026	96.96	0.125	90.34
2	0.016	98.11	0.121	90.62
3	0.009	98.93	0.067	94.82
4	0.017	98.05	0.122	90.6
5	0.866	0	1.294	0

The application of treatments for the lesion nematode *Pratylenchus* sp., presented a significant control effectiveness ($p < 0.05$) that fluctuated from 87.47 to 92.87% (Table 3), treatments that reduced the population density in a range of 0.067-0.125 times the initial population, highlighting with greater efficiency (94.82%) treatment 3 (Nemaxxion XT Plus 6 L ha^{-1}) (Table 4).

The control treatment without nematode control did not present a reduction in the population (Table 3) (gl= 8, 24; F= 416.54; p< 0.0001; R^2= 0.99; C.V.= 4.35), with a population increase of 1.294 times (Table 4), increasing 13 more nematodes to the starting population (Table 2).

Based on the results, the use of the Nemaxxion XT Plus product based on plant extracts and organic components showed high efficiency for the control of *M. incognita* and *Pratylenchus* sp., reducing the density of the populations in the coffee tree crop; whose effect is similar to the organophosphate product Nemacur® 400 CE, which suggests that the product of biological origin can be used as a control alternative for these nematodes.

Various authors have pointed out the importance and effectiveness of using ecological alternatives for the management of nematodes, mainly plant extracts, which allows the development of a more profitable and non-polluting agriculture (Parada and Guzmán, 1997; Dama, 2002; Jothi *et al*., 2004; Vinueza *et al*., 2006; Zakaria *et al*., 2013; Garrido *et al*., 2014).

Conclusions

Nemaxxion XT Plus is effective for the control of the root-knot nematode *Meloidogyne incognita* and the lesion nematode *Pratylenchus* sp., in coffee cultivation, in addition to its organic and environmentally friendly characteristics it can be used in any agricultural production system, presenting itself as an alternative for a more sustainable agriculture.

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