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RESEARCH ARTICLE

Resistance of Conilon coffee cultivar Vitoria Incaper 8142 to *Meloidogyne paranaensis* under field conditions

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Abstract

Meloidogyne paranaensis is responsible for considerable losses in coffee production. Because of the distribution of this species in the main *Coffea arabica* producing regions, there is a need for management practices to ensure the sustainability of coffee production. In this work, we evaluated the agronomic performance of resistant clones of the Conilon coffee cultivar Vitoria Incaper 8142 in areas infested by *M. paranaensis* in the west region of Minas Gerais, Brazil. Clones 2V, 3V, and 6V presented the lowest number of nematodes per gram of roots and were considered resistant to *M. paranaensis*. All other clones were considered tolerant to this nematode, and one had good vegetative growth but allowed nematode reproduction. Clones of Vitoria Incaper 8142 of *C. canephora* represent an alternative to coffee production in areas infested by *M. paranaensis* including areas traditionally cultivated with *C. arabica*.

Keywords: Root-knot nematodes; *Coffea canephora*; Genetic resistance

Introduction

Root-knot nematodes (RKNs) are recognized as major agricultural pathogens, causing expressive losses in many crops (Jones et al., 2013). Among several species that parasitize coffee plants, *Meloidogyne paranaensis* (Carneiro et al., 1996) is especially important, hindering plant growth and causing losses of approximately 35% in yield (Lopez-Lima et al., 2015). The damage can be even higher depending on the level of attack and on the genetic variability within nematode population (Santos et al., 2018).

M. paranaensis is widely spread in *Coffea arabica* and *C. canephora* coffee growing areas in Brazil and other Latin American countries (Barros et al., 2011; Barros et al., 2014; Villain et al., 2013). The alarming rise in the occurrence of this nematode in Minas Gerais State (Castro et al., 2003; Castro et al., 2008; Salgado et al., 2015), the largest *C. arabica*-producing state in Brazil, requires management practices to ensure the sustainability of coffee production in these areas. The use of resistant cultivars is the best way to cultivate coffee plants in soils infested by RKNs. While most of *C. arabica* cultivars are susceptible to *M. paranaensis*, *C. canephora* shows genetic variability in their resistance to this nematode species (Bertrand et al., 2000; Fatobene et al., 2018).

Some clones of Conilon coffee cultivar Vitoria Incaper 8142 of *C. canephora* exhibited multiple resistance to *M. exigua*, *M. incognita*, and *M. paranaensis* (Lima et al., 2015) under controlled conditions. Because Conilon coffee is recommended for regions at low altitudes, the goal of this

study was to evaluate the agronomic performance of resistant clones of Conilon coffee cultivar Vitoria Incaper 8142 in areas infested by *M. paranaensis* in the west region of Minas Gerais.

Material and Methods

In this study, we evaluated five coffee clones 2V, 3V, 6V, 7V, and 13V resistant to *M. paranaensis*, and the susceptible clone 12V, previously screened by Lima et al. (2015). Cultivar Catuaí Vermelho IAC 99 of *C. arabica* was used as a susceptible control. Clonal seedlings were formed from cuttings derived from orthotropic branches (OB) in polyethylene bags with commercial substrate (Fonseca et al., 2007).

The experiment was conducted for 4 years in Piumhi MG, Brazil (20°25′28″S; 46°1′10″W; 812 m a.s.l.), in a field previously cultivated with *C. arabica* and infested by *M. paranaensis* esterase phenotype P1 (Carneiro and Almeida, 2001). The soil texture was 61% clay, 28% silt, and 11% sand. The average air temperature and rainfall during the growing seasons were 20.5 °C and 1588 mm in 2011/2012; 21.1 °C and 1417 mm in 2012/2013; 21.0 °C and 924 mm in 2013/2014; and 21.3 °C and 1259 mm in 2014/2015. Clonal seedlings of the Vitoria Incaper 8142 and susceptible control Catuaí Vermelho IAC 99 were planted in a randomized block design with five replicates, that is, plots of seven plants, spaced 3.5 m between rows and 0.7 m between plants, in December 2012.

The infective population of *M. paranaensis* in the soil was evaluated twice: at the beginning of the experiment (Biotest 1) and at the first coffee harvest in June 2015 (Biotest 2). Soil samples from three equidistant spots in the plot were collected, mixed, and placed in 550 cm^3 plastic pots to grow the susceptible tomato cultivar Santa Clara. Then, the population of *M. paranaensis*, eggs + J2, was quantified in tomato roots after 60 days. Extraction of nematodes was conducted according to Hussey and Barker (1973), adapted by Bonetti and Ferraz (1981). The population of *M. paranaensis* (eggs + J2) was also evaluated in roots of coffee clones. Four subsamples of roots were collected in equidistant spots at 20 cm from the central axis of the plant and a depth of 30 cm, both perpendicular to the direction of the crop line. The samples were collected during the first coffee harvest in June 2015. Again, nematodes were extracted according to Hussey and Barker (1973), adapted by Bonetti and Ferraz (1981) and counted using a Peter's slide under a biological microscope. Population density was expressed as the number of eggs + J2 per gram of roots (NEM g^{-1}).

Vegetative traits like plant height (PH), in centimeters from the stem base to the apex of the plants, number of OB, stem diameter (SD), in centimeters measured at the stem base 10 cm above the ground, and canopy diameter (CD), in centimeters measured at 50 cm above the ground, were evaluated 48 months after planting. The vegetative vigor (VV) was evaluated according to the scale proposed by Carneiro (1995), where the score 5 means plants with excellent VV while the score 0 indicates extremely depleted or dead plants. Plants that scored from 0 to 2 were considered susceptible, and from 3 to 5 were considered tolerant and/or resistant. They were also evaluated for the percentage of dead plants (%DP). Initial reproductive development was evaluated through fruit yield (FY) in liters and the percentage of fruit with at least one empty locule (%FEL). Fruit ripening was evaluated through the percentage of green fruits (%GF).

An analysis of variance (ANOVA) was performed using the 'agricolae' package of the R statistical software (R Core Team, 2016; Mendiburu, 2015). Data were transformed when they did not fit the assumptions of the ANOVA. The clone means were compared by Tukey's test using the 'HSD.test' function from the 'agricolae' package ($p < 0.05$).

Results

The presence and infectivity of *M. paranaensis* in the experimental field were confirmed in both biotests (Table 1). No significant differences ($p > 0.05$) were obtained between the number of

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Table 1. Average number[1] of *Meloidogyne paranaensis* (eggs + J2) on roots of tomato plants grown in soil sampled in clone plot biotests

	Eggs + J2	
Treatments	Biotest 1	Biotest 2
2V	604.5 aA	129.0 aB
3V	158.6 aA	26.5 aA
6V	568.5 aA	570.3 aA
7V	206.1 aA	168.1 aA
12V	183.3 aA	335.3 aA
13V	140.9 aA	270.5 aA
CV IAC 99[2]	182.9 aB	2862.5 aA

[1]Means followed by the same capital letter in rows and by the same lowercase letter in columns do not differ at the 0.05 probability level according to the Tukey's test.
[2]Susceptible control *Coffea arabica* cultivar Catuaí Vermelho IAC 99.

Table 2. Number of eggs + J2 of *Meloidogyne paranaensis* per gram of root (NEM g^{-1}), vegetative vigor (VV), and percentage of dead coffee plants (%DP)

Treatments	NEM g^{-1}	VV[1]	%DP
2V	7.0 a	5.0 a	22.9
3V	2.4 a	4.6 a	25.7
6V	10.9 a	4.6 a	25.7
7V	20.9 ab	5.0 a	28.6
12V	47.1 ab	5.0 a	28.6
13V	49.7 ab	3.4 a	37.1
CV IAC 99[2]	315.9 b	0.8 b	62.9

For NEM g^{-1} and VV, means followed by the same letter in columns do not differ at the 0.05 probability level according to the Tukey's test.
[1]Vegetative vigor scores according to the scale of Carneiro (1995).
[2]Susceptible control *Coffea arabica* cultivar Catuaí Vermelho IAC 99.

nematodes in clone plots in both individual and joint variance analyses, indicating that the infestation of nematodes was homogeneous in the field. This condition was essential to the avoidance of erroneous evaluation of clone resistance. Significant effects were observed for the interaction years × clones in the joint ANOVA. There was a decrease in the nematode population in clone 2V, and an increase in the nematode population in the susceptible control Catuaí Vermelho IAC 99.

Clones 2V, 3V, and 6V showed low nematode population density and good VV (Table 2). Clones 7V and 13V showed intermediate means of NEM g^{-1} but also presented high scores of VV. Clone 12V, previously classified as susceptible to *M. paranaensis*, presented vegetative growth and FY similar to those of the other Conilon coffee clones. As expected, the susceptible control Catuaí Vermelho IAC 99 had the largest NEM g^{-1} and consequently the lowest VV and high %DP.

On average, the vegetative traits in clones of Vitoria Incaper 8142 were significantly higher than in the susceptible control Catuaí Vermelho IAC 99 (Table 3). While some of these differences could be influenced by the resistance to nematodes, others were caused by botanical characters intrinsic to each species, such as the higher number of OB in *C. canephora* clones. Clones of Vitória Incaper 8142 showed higher FY and later ripening than the susceptible control Catuaí Vermelho IAC 99 (Table 4).

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Table 3. Stem diameter (SD), canopy diameter (CD), number of orthotropic branches (OB), and plant height (PH) of *Coffea canephora* clone cultivar Vitoria Incaper 8142 and *Coffea arabica* cultivar Catuaí Vermelho IAC 99 in a field infested by *Meloidogyne paranaensis*

Treatments	SD (cm)	CD (cm)	OB	PH (cm)
2V	10.8 a	94.8 a	3.7 b	81.2 a
3V	11.4 a	99.9 a	2.4 cd	71.9 ab
6V	10.4 a	98.4 a	3.5 bc	73.4 ab
7V	10.1 a	89.0 a	5.6 a	74.1 ab
12V	11.5 a	102.8 a	3.4 bc	81.6 a
13V	9.8 a	93.6 a	3.1 bc	67.9 bc
CV IAC 99[1]	7.2 b	67.9 b	1.5 d	58.9 c

Means followed by the same letter in columns do not differ at the 0.05 probability level according to the Tukey's test.
[1]Susceptible control *Coffea arabica* cultivar Catuaí Vermelho IAC 99.

Table 4. Average fruit yield (FY), percentage of green fruits (%GF), and percentage of fruits with at least one empty locule (%FEL) for clones of *Coffea canephora* cultivar Vitoria Incaper 8142 and *Coffea arabica* cultivar Catuaí Vermelho IAC 99 in a field infested by *Meloidogyne paranaensis*

Clones	FY	%GF	%FEL
2V	18.2 a	77.6 a	6.0
3V	9.2 ab	58.0 ab	12.8
6V	15.0 a	28.2 bc	11.2
7V	11.2 ab	69.2 a	5.2
12V	12.2 ab	27.2 bc	13.6
13V	9.8 ab	83.2 a	12.8
CV IAC 99[1]	0.2 b	5.2 c	9.2

For FY and %GF, means followed by the same letter in columns do not differ at the 0.05 probability level according to the Tukey's test.
[1]Susceptible control *Coffea arabica* cultivar Catuaí Vermelho IAC 99.

Discussion

The clones 2V, 3V, 6V 7V, and 13V of Vitoria Incaper 8142, classified as resistant to *M. paranaensis* under controlled conditions (Lima et al., 2015), also showed good performance in the field infested by *M. paranaensis*. The clones 2V, 3V, 6V, and 7V were also resistant to *M. exigua* (Lima et al., 2015), a valuable feature considering the widespread nature of this species within coffee plantations. In addition to the survival and good fitness of plants, integrated management of nematodes must consider the ability of plants to reduce the nematode population in fields as the extensive use of mechanization may promote the dissemination of nematodes. For this reason, clone 12V should not be planted in soils infested by *M. paranaensis*, despite its good performance.

The low FYs of *C. arabica* Catuaí Vermelho IAC 99 in the first harvest were also observed by Carvalho et al. (2017) in areas infested by *M. paranaensis*. On the other hand, yields of clones were not affected by nematode parasitism, even in a distinct edaphoclimatic region at an altitude about 800 m. In fact, clones of the Conilon coffee-like cultivar Vitória Incaper 8142 were recommended for regions with average annual air temperatures between 22 and 26 °C and altitudes below 650 m (Taques and Dadalto, 2017).

Most studies evaluating the performance of coffee genotypes cultivated in soil infested by *Meloidogyne* spp. were conducted under controlled conditions. Although requiring more time, the evaluation of genotypes in the field is extremely important for coffee breeding programs because the evaluation is more reliable (Oliveira et al., 2011) and the selection is more efficient.

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The use of coffee cultivars resistant to nematodes and adapted to specific environmental conditions can effectively contribute to the improvement of plant performance in infested areas, increasing yield, reducing production costs, and ensuring greater competitiveness and sustainability of coffee production (Carvalho et al., 2017).

Genetic control of nematodes has been implemented with relative success in coffee breeding. The first strategy adopted by breeding programs was the development of resistant *C. canephora* rootstocks Apoatã IAC 2258 (Gonçalves et al., 1996) and Nemaya (Bertrand et al., 2000). Both are resistant to *M. exigua* but with different levels of resistance segregation to *M. incognita* and *M. paranaensis*. A more recent study identified clones of *C. canephora* with multiple resistance to *M. incognita* and *M. paranaensis* (Fatobene et al., 2018) that could be used in the breeding of new rootstocks or ungrafted cultivars of *C. canephora*. Currently, some ungrafted cultivars of *C. arabica* resistant to RKNs are available, such as IPR 100 resistant to *M. paranaensis* (Sera et al., 2007) and to *M. exigua* (Rezende et al., 2017), and IAC 125 RN (Fazuoli et al., 2018) and IAPAR 59 (Salgado et al., 2005) resistant to *M. exigua*.

Considering resistance to RKNs and the increasing demand for 'Robusta' and 'Conilon' coffees in the international market, clones 2V, 3V, 6V, and 7V of Vitoria Incaper 8142 of *C. canephora* represent alternatives to coffee production in areas infested by *M. paranaensis*, as well as in areas traditionally cultivated with *C. arabica*.

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