Four new grape hybrids resistant to *Meloidogyne javanica*

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Abstract – Root-knotting nematodes are found in vineyards and are associated with grapevine decline. This study aimed to evaluate the resistance of four new hybrids, obtained by crossing *Vitis labruscana* and *Vitis rotundifolia* (IBRE 421, IBBT 481, IBRK 504 and IBMG 631), to *Meloidogyne javanica.* This study also evaluated three rootstocks (IAC 766, Paulsen 1103, VR 043-43), widely used and showing resistance levels to *M. javanica*. The plants were obtained from cuttings, then grown in pots, and inoculated with a population of *M. javanica*. One year after inoculation, the plants presented no galls on their fine roots. The root nematodes were extracted and quantified. The reproduction factor of *M. javanica* in all genotypes was close to zero. The tested hybrids are not good hosts for the multiplication of *M. javanica*. Thus, these new hybrids can be used as a genetic control strategy of *M. javanica.*

Index terms: *Vitis labruscana; Vitis rotundifolia;* root-knot nematode.

Quatro novos híbridos de videira resistentes a *Meloidogyne javanica*

Resumo - Os nematoides formadores de galhas são encontrados em vinhedos e estão associados ao declínio das videiras. O objetivo do trabalho foi avaliar a resistência de quatro novos híbridos obtidos pelo cruzamento entre *Vitis labruscana* e *Vitis rotundifolia* (IBRE 421, IBBT 481, IBRK 504 e IBMG 631) quanto a resistência a *Meloidogyne javanica*. Foram avaliados também 3 porta-enxertos (IAC 766, Paulsen 1103, VR 043-43) amplamente utilizados e que apresentam níveis de resistência a *M. javanica*. As plantas foram obtidas por estaquia, cultivadas em vasos e inoculadas com uma população de *M. javanica*. Um ano após a inoculação, as plantas não apresentavam galhas em suas raízes finas. Os nematoides nas raízes foram extraídos e quantificados. O fator de reprodução de *M. javanica* em todos os genótipos foi próximo de zero. Os híbridos testados não são bons hospedeiros para a multiplicação de *M. javanica*. Assim, esses novos híbridos podem ser utilizados como estratégia para o controle genético de *M. javanica*.

Termos de indexação: *Vitis labruscana; Vitis rotundifolia;* nematoide das galhas.

Brazilian grape production in 2019 was estimated at 1.4 million tons grown on 74.2 thousand ha^{-1}, with an average productivity of 19.4 tons ha^{-1} (IBGE, 2021). The south of the country concentrates about 60% of the national grape production (IBGE, 2021). The Brazilian viticulture is diverse, including production chains formed by fine grape cultivars of the species. Thus, development of new grape genotypes is necessary for the consumer market, adapting them to edaphoclimatic conditions, with high productivity and resistance to the main pests and diseases.

In Brazil, nematodes in the genus *Meloidogyne*, known as the root-knot nematodes, is the major economic important group, and the *Meloidogyne javanica* is frequently found in grapevine roots (SOMAVILLA, 2011). In sandy soils and in mild temperatures, they cause great damage to the culture (TÉLIZ et al., 2007). The increase of the root-knot nematode population in the grapevine roots affects plant growth, productivity, evolving to the decline of grapevines. Eradication of nematodes in the soil is practically impossible, and the growers must constantly manage the nematode population to maintain it at levels that have little effect on production. Chemical control is frequently used, but it is associated with increased production costs due to the numerous applications necessary, in addition to environmental and human health impacts. In this context, the use of rootstocks resistant to nematodes is an effective and economical strategy for the control of root-knot nematodes. However, information on the resistance of grapevine rootstocks to nematodes in Brazil is scarce. Thus, this study aimed to evaluate four new grapevine rootstocks hybrids for resistance to *M. javanica*.

The cuttings of the rootstocks IAC 766 (*Vitis caribaea* x (*Vitis riparia* x (*Vitis rupestris* x *Vitis cordifolia*))), Paulsen 1103 (*Vitis berlandieri* x *V. rupestris*), and VR 043-43 (*V. vinifera* x *V. rotundifolia*); and the hybrids IBRE 421, IBBT 481, IBRK 504, and IBMG 631

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(*Vitis labruscana* x *V. rotundifolia*) were obtained from matrix plants grown at the Experimental Farm Canguiri in the municipality of Pinhais, PR, Brazil. Bare-root cuttings were transplanted into 2 dm^3 aluminum pots, containing OXISOL and sand in the proportion of 1:1 (v v^{-1}), previously autoclaved at 121°C for 1 h. Eight months later, the cuttings with the soil were transplanted into 8 dm^3 plastic pots. The experiment was carried out in a randomized block design with seven genotypes and three replicates, each replication was a plant. Tomato seedlings of the cultivar Santa Clara were used as control to test the inoculum viability

The *M. javanica* inoculum was extracted from tomato plants using the sucrose centrifugal flotation technique with caulin (COOLEN & D'HERDE, 1972). Nematodes were counted in a Peters chamber under an optical microscope at 40x magnification. The nematodes were counted in all quadrants of the Peters chamber and multiplied by the total root fresh mass, thus obtaining the final population of the sample. One milliliter of suspension with 5,000 eggs (initial population — IP) was deposited in each plant. After 370 days, the root system was evaluated to check for the presence of galls. Samples of 10 g of roots were collected from each plant to extract the nematodes, using the sucrose centrifugal flotation technique with caulin (COOLEN & D'HERDE, 1972). Nematodes were counted in the same way as for inoculum production. The final population (FP) was estimated by multiplying the number of nematodes per gram of roots to the fresh mass of roots. The reproduction factor (RF) was calculated by dividing the FP by the IP of nematodes (OOSTENBRINK, 1966). The gall index (GI) was determined considering: 0 = no gall, 1 = 1-2 galls, 2 = 3-10, 3 = 11-30, 4 = 31-100, 5 > 100 (TAYLOR & SASSER, 1978). The data were subjected to analysis of variance and Tukey's test ($p<0.05$) in the ExpDes.pt package in R software.

Regarding mean gall index, Table 1 shows the number of nematodes per gram of roots and the RF for the grapevine genotypes. No galls were found in the roots of the grapevine genotypes. The number of nematodes in the genotypes was less than 12 nematodes per gram of roots. Consequently, the reproduction factor of *M. javanica* for all tested grapevine genotypes was close to zero. Tomato plants showed intense gall formation (Figure 1), presenting a high reproduction factor (RF=68.6). This indicates that the inoculum was viable, giving reliability to the results that showed that the tested hybrids are not good hosts for the multiplication of *M. javanica*.

The IAC-766 rootstock is not a suitable host for *M. javanica*, confirming the observations by Moura et al. (2014). This rootstock is not a suitable host for *M. incognita* (MOURA et al., 2014), *Meloidogyne enterolobii* (FREITAS et al., 2017) and *Meloidogyne arenaria* (SOMAVILLA, 2011). Moreover, the VR 043-43 rootstock is not a suitable host for *M. javanica*. This rootstock was developed to be resistant to *Xiphinema index* (FERRIS et al., 2012).

The hybrids IBBT 481, IBRK 504, IBMG 631 and IBRE 421, obtained by interspecific hybridization between the cultivar Isabel of the American species *V. labruscana* and cultivars of muscadine grapevines of the species *V. rotundifolia* (SCHUCK et al., 2011), showed reproduction factors to *M. javanica* close to zero. Cultivars of the species *V. rotundifolia* are a source of resistance to several species of nematodes (FERRIS et al., 2012).

The use of resistant rootstocks is frequent. However, resistance superseded by virulent populations of *M. arenaria* and *M. incognita* has been reported in California (FERRIS et al., 2012), and *M. javanica* in Australia (SMITH et al., 2017).

When considering the importance of nematodes regarding geographic distribution and difficulty in control, genetic control of nematodes via resistant rootstocks constantly needs new grapevines genotypes adapted to edaphoclimatic conditions, and that can replace rootstocks with superseded resistance or inferior agronomic characteristics. Thus, the hybrids IBRE 421, IBBT 481, IBRK 504 and IBMG 631 are not good hosts for the multiplication of *M. javanica* and appear as new options to be used in the control of *M. javanica* in grapevines.

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Table 1. Reaction of grapevine rootstock hybrids to Meloidogyne javanica grown for 370 days after inoculation, based on gall index (GI), number of nematodes per gram of roots (N) and the reproduction factor (RF)

Tabela 1. Reação de híbridos de porta-enxertos de videira a *Meloidogyne javanica* cultivados por 370 dias após a inoculação, com base no índice de galhas (IG), número de nematóides por g de raízes (N) e fator de reprodução (FR)

Genotypes	IG	N	FR
IAC-766	0.00 ± 0.00 ^{ns}	1.22 ± 0.57 ^{ns}	0.04 ± 0.02 ^{ns}
Paulsen 1103	0.00 ± 0.00	11.51 ± 8.24	0.14 ± 0.08
IBRE 421	0.00 ± 0.00	6.77 ± 5.80	0.08 ± 0.07
IBBT 481	0.00 ± 0.00	4.23 ± 3.43	0.09 ± 0.10
IBRK 504	0.00 ± 0.00	5.14 ± 1.80	0.03 ± 0.02
IBMG 631	0.00 ± 0.00	8.82 ± 4.13	0.21 ± 0.12
VR 043-43	0.00 ± 0.00	5.63 ± 3.45	0.18 ± 0.13

^{ns} Means did not differ significantly by Tukey's test, $p<0.05$.

^{ns} Médias não diferiram significativamente pelo teste Tukey, $p<0.05$.

Figure 1. Roots of vine rootstock genotypes (resistant) and tomato roots (susceptible) infested with Meloidogyne javanica. A) IAC-766; B) Paulsen 1103; C) IBRE 421; D) IBBT 481; E) IBRK 504; F) IBMG 631; G) VR 043-43; and, H) Tomato of the cultivar Santa Clara with detail of the female of M. javanica inside a gall
Photo: Marlon Henrique Hahn
Figura 1. Raízes de genótipos de porta-enxertos de videira (resistente) e raízes de tomateiro (suscetível) infestadas com *Meloidogyne javanica*. A) IAC-766; B) Paulsen 1103; C) IBRE 421; D) IBBT 481; E) IBRK 504; F) IBMG 631; G) VR 043-43; e, H) Tomate da cultivar 'Santa Clara' com detalhe da fêmea de *M. javanica* no interior de uma galha.
Foto: Marlon Henrique Hahn

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