

Anthelmintic activity of plant aqueous extracts against *Panagrellus redivivus* in vitro

Atividade anti-helmíntica de extratos aquosos de plantas contra *Panagrellus redivivus* in vitro

Cleonice Lubian^{1*} , Danielle Dutra Martinha² , Roberto Portz² , Manoel Penachio Gonçalves² , Sabrina Holz³ , Wesler Luiz Marcelino³ , Ana Claudia Constantino Nogueira⁴ , Renata Mori Thome⁵ , Vivian Carré Missio² , Juliano Cordeiro² , Lorraine Tomim Feroldi² 

ABSTRACT: Control of phytonematodes is very hard and requires a combination of techniques to succeed. Alternative control through plant extracts may result in the discovery of nematicide substances. Research aimed at evaluating the effect of 33 plants submitted to aqueous extraction against *Panagrellus redivivus* in vitro. Concentrations were prepared at 1.25, 2.5, 5, 10, and 20%. Monitoring happened at 0, 6, 12, 24 and 30 hours after preparation. Counting considered dead nematodes subtracted from alive ones. Juveniles were also counted, and extract efficiency was expressed in percentage of control or stimuli. Data were submitted to variance analysis. Significant results got with the Scott-Knott test (5%), and multiple linear regression analysis. Extracts were observed acting as controllers, but also as stimulators to nematode reproduction. The best controlling performance was set by *Carica papaya* (-66% at 20%; -33.7% at 10%), *Euphorbia milii* (-37% at 20%), *Psychotria carthagenensis* (-25.5% at 2.5%), *Clusia variegata* (-22% at 20%), and *Zamioculcas zamiifolia* (-21.5% at 20%). Stimulator extracts were *Mentha villosa* at 10% (+148%) and 2.5% (+131.5%), followed by *Aloe vera* (+123% at 5%), *Schinus molle* (+112.5% at 10%), *Schefflera arboricola* (+93.5% at 5%), *C. variegata* (+89% at 5%), and *S. molle* (+88% at 5%). Some extracts kept population stable throughout the experiment, presenting lower control indexes. Besides an additive effect, there was an individual influence of concentration or time on control.

KEYWORDS: pesticide potentiality; nematicide compounds; phytopathology.

RESUMO: O controle de fitonematoides é muito difícil e requer uma combinação de técnicas para ter sucesso. O controle alternativo via extrato vegetal pode resultar na descoberta de substâncias nematicidas. Esta pesquisa objetivou avaliar o efeito de 33 plantas submetidas à extração aquosa contra *Panagrellus redivivus* in vitro. As concentrações foram preparadas a 1,25; 2,5; 5; 10; e 20%. O monitoramento ocorreu em 0, 6, 12, 24 e 30 horas após a preparação. Para a contagem, foram considerados nematoides mortos subtraídos dos vivos. Nematoides jovens também foram contados, e a eficiência dos extratos foi expressa em porcentagem de controle ou de estímulo. Os dados foram submetidos à análise de variância. Resultados significativos foram analisados pelos testes de Scott-Knott (5%) e análise de regressão múltipla. Foram observados extratos agindo como controladores, bem como estimuladores da reprodução de nematoides. A melhor performance de controle foi obtida por *Carica papaya* (-66% a 20%; -33,7% a 10%), *Euphorbia milii* (-37% a 20%), *Psychotria carthagenensis* (-25,5% a 2,5%), *Clusia variegata* (-22 a 20%) e *Zamioculcas zamiifolia* (-21,5% a 20%). Os extratos estimuladores foram *Mentha villosa* a 10% (+148%) e 2,5% (+131,5%), seguido por *Aloe vera* (+123% a 5%), *Schinus molle* (+112,5% a 10%), *Schefflera arboricola* (+93,5% a 5%), *C. variegata* (+89% a 5%) e *S. molle* (+88% a 5%). Alguns extratos mantiveram a população estável durante todo o experimento, apresentando menores índices de controle. Além do efeito aditivo houve uma influência individual da concentração e do tempo no controle.

PALAVRAS-CHAVE: potencial pesticida; componentes nematicidas; fitopatologia.

¹Universidade Estadual do Oeste do Paraná – Marechal Cândido Rondon (PR), Brazil

²Universidade Federal do Paraná – Palotina (PR), Brazil

³Escola Superior de Agricultura “Luiz de Queiroz”, Universidade de São Paulo – Piracicaba (SP), Brazil

⁴Centro Universitário Integrado – Londrina (PR), Brazil

⁵Universidade Estadual de Londrina – Londrina (PR), Brazil

*Corresponding author: cleo.lubian@gmail.com

Received on: 05/25/2018. Accepted on: 07/29/2019

INTRODUCTION

Plants are affected by biotic and abiotic factors, which leads to reduced productivity levels. Among biotic factors, diseases caused by phytoparasitic nematodes compromise root functions in soil, as water and nutrients uptake and plant support (CAMPOS et al., 2011).

Meloidogyne is the most important genus followed by *Heterodera*, *Globodera*, and *Pratylenchus* (JONES et al., 2013). Nematoda phylum contains over 27,000 species already described (QUIST et al., 2015), including free-living nematodes and those that affect animals. Related to phytonematodes, over 4,100 species were registered, causing damage in crops, corresponding to roughly 80 billion dollars per year (DE ALMEIDA ENGLER; FAVERY, 2011).

There are few viable techniques to control nematodes in croplands, resorting on chemical method, although it presents a high toxicity level (NTALLI; CABONI, 2017). In this context, alternative methods have been deeply studied as alternative methods, especially considering environmental concerns and human health, focused on decreasing nematicides use (DIAS et al., 2016).

Plant extract is practiced since 1972 (NAKASHIMA; SHIMIZU, 1972), and stands out because of its potential molecules with nematicidal activity, derived from secondary metabolism. Both aqueous and alcoholic plant extract were studied against nematodes (TARIQ et al., 2009). Many oils not only repeal plagues, but also present contact and fumigant action on nematodes (ISMAN, 2000). Nematicide oils are especially important in small areas (GARDIANO et al., 2009).

Given the easy handling of *Panagrellus redivivus* in laboratory, research aimed at testing nematicidal activity of aqueous extract from 33 plant species against *P. redivivus*. We considered cultural knowledge to hypothesize that new plants may contain nematicidal substances.

MATERIAL AND METHODS

Samples of 33 plant species were collected during spring season. Some plants have their athelmintic activity already described in literature (Table 1).

Plants were sent to the Phytopathology Laboratory of *Universidade Federal do Paraná* – Setor Palotina. Leaves were cut off for extract preparing. In the case of *Carica papaya*, we used seeds. Plant parts were weighted (40 g) and mixed in 60 mL of distilled water in a blender for three minutes, then filtrated on gauze. The resulting liquid was stored into test tubes and frozen at -20°C for two weeks. For evaluation, such tubes were defrosted naturally for 24 hours. Next, the liquid was centrifuged for five minutes at 4,000 rpm applied for decanting substances elimination.

Dilutions were prepared just before adding nematodes, obeying a serial dilution composed by 1 mL directly pipetted from each extract placed into small Petri plates (60 × 15 mm), jointly with 1 mL of distilled water, representing the first dilution of 20% of extract concentration. The following dilutions were performed in the same pattern, at 10, 5, 2.5, and 1.25% (w/v), with four repetitions each. Control treatment was composed of distilled water. The experimental design was in a completely randomized set in room temperature ($25 \pm 2^{\circ}\text{C}$).

Panagrellus redivivus were maintained in a creamy mixture of oat flour and distilled water. Their collection comprised nematodes climbing the boundaries of the container they were into. In sequence, nematodes were washed to be free from flour particles. Population was calibrated with water into a Bequer to enable the medium collection of 10 individuals in 20 μL of water drew up by electronic micropipette. This water volume was completed with 80 μL of distilled water, the final volume.

For nematode death (non-motile) evaluation, the initial number of nematodes added into Petri plates was identified on the top of each lid to allow a precise following counting in a stereo microscope. In addition, newborn nematodes (juveniles) were also counted to determine whether extracts would stimulate *P. redivivus* reproduction.

Data were registered in spreadsheets for posterior percentage calculation to set how effective or which stimulator each extract it was. Evaluation criterion considered the balance of alive nematodes at all evaluation times, of which values were subtracted from the initial ones.

The first evaluation occurred after adding nematodes to check any possible immediate effect of extracts on their behavior, considering the reaction pattern, especially as to how fast they moved. Next counting was conducted after 6, 12, 24, and 30 hours of interaction with each extract.

Data were submitted to variance analysis (ANOVA). In cases of significant results, Scott-Knott media test was applied at 5% of error probability ($p = 0.05$) to group extracts, according to their similarity. Extracts presenting distinguished effects were analyzed with the linear regression test ($p = 0.05$), which was applied individually to time and dose factors to determine their particular influence on nematodes. As the dependent variable (alive nematodes) was explained by dose and time combined, the multiple regression test ($p = 0.05$) was applied to all extracts to describe, mathematically, the relation between independent and dependent variables. The usefulness of regression prediction with the equation was set by determination coefficient. Tests were performed with SISVAR 5.6[®] program (FERREIRA, 2011).

RESULTS

ANOVA presented significance for time, dose, extract, extract* dose and extract* time (data not presented). To these parameters, Scott-Knott media test was applied at 5% of probability.

Concerning data presentation, negative signal before percentage values determines nematodes control, whereas the absence of signal represents stimulation on nematode reproduction.

At 0 hour, a maximum stimulation and reduction of 10 and -14%, respectively, was found (Table 2). However, the number expressed at this time refers to the real number of nematodes added to each extract; therefore, it is possible for eight nematodes to have been counted for *Agave angustifolia*, for example. Regardless of that, there was no significant

difference at 0 hour, revealing no readily effects of any extract. Fluctuations as from six hours reveals the real effects of extracts. The highest control percentage was -17.08%, reached for *Peschiera fuchsiaefolia* at 30 hours; the highest stimulation, 170.42%, was caused by *Mentha villosa* at 30 hours.

Extracts varied on effectiveness and many of them kept the nematode population stable throughout the experiment, such as *Garcinia gardneriana*, *Psychotria carthagenensis*, *Bauhinia forficata*, *P. fuchsiaefolia*, *Genipa americana*, *Ligustrum lucidum*,

Table 1. Common names, scientific names and botanical family of collected plants.

Common name in Portuguese	Family	Scientific name
Agave (Or)	Asparagaceae	<i>Agave angustifolia</i> Haw. (1812)
Babosa (M)	Asphodelaceae	<i>Aloe vera</i> (L.) Burm. f. (1768)
Bacupari (A)	Clusiaceae	<i>Garcinia gardneriana</i> (Planch. & Triana) Zappi (1993)
Cana-do-brejo (M)	Costaceae	<i>Costus spicatus</i> (Jacq.) Sw. (1788)
Carqueja (M)	Asteraceae	<i>Baccharis trimera</i> (Less.) DC. (1836)
Cedro (A)	Meliaceae	<i>Cedrela fissilis</i> Vell. (1825 [1829])
Cheflera (Or)	Araliaceae	<i>Schefflera arboricola</i> Hayata (1916)
Aroeira-salsa (A)	Anacardiaceae	<i>Schinus molle</i> L.
Clusia (Or)	Clusiaceae	<i>Clusia variegata</i>
Comigo-ninguém-pode (Or)	Araceae	<i>Dieffenbachia seguine</i> (Jacq.) Schott (1829)
Coroa-de-cristo (Or)	Euphobiaceae	<i>Euphorbia milii</i> Des Moul. (1826)
Embaúba (A)	Urticaceae	<i>Cecropia pachystachya</i> Trécul. (1847)
Erva-de-rato (W)	Rubiaceae	<i>Psychotria carthagenensis</i> Jacq. (1760)
Espada-de-São-Jorge (Or)	Asparagaceae	<i>Sansevieria trifasciata</i> Prain (1903)
Figo (S)	Moraceae	<i>Ficus carica</i> L. (1753)
Forquilha (M)	Apocynaceae	<i>Peschiera fuchsiaefolia</i> (A. DC.) Miers
Guiné (M)	Petiveriaceae	<i>Petiveria alliacea</i> L. (1753)
Hortelã (M)	Lamiaceae	<i>Mentha villosa</i> Huds. (1778)
Jenipapo (A)	Rubiaceae	<i>Genipa Americana</i> L. (1759)
Ligustre (A)	Oleaceae	<i>Ligustrum lucidum</i> W. T. Aiton (1810)
Losna (M)	Asteraceae	<i>Artemisia absinthium</i> (Mill.) DC. (1815)
Mamão (A)	Caricaceae	<i>Carica papaya</i> L. (1753)
Mandioca-brava (S)	Euphobiaceae	<i>Manihot esculenta</i> Crantz (1766)
Maria-preta (W)	Solanaceae	<i>Solanum americanum</i> Mill. (1768)
Mulungu (Or/M)	Fabaceae	<i>Erythrina verna</i> Vell. (1825)
Noni (A/M)	Rubiaceae	<i>Morinda citrifolia</i> L. (1753)
Pau-d'alho (A)	Petiveriaceae	<i>Gallsia integrifolia</i> (Spreng.) Harms (1934)
Pata-de-vaca (A)	Fabaceae	<i>Bauhinia forficata</i> L. (1753)
Pinhão-roxo (A)	Euphobiaceae	<i>Jatropha gossypifolia</i> L. (1866)
Poejo (M)	Lamiaceae	<i>Mentha</i> sect. <i>pulegium</i> Coss. & Germ. (1845)
Sabugueira (M)	Adoxaceae	<i>Sambucus nigra</i> L. (1753)
Trombetarosa (Or)	Solanaceae	<i>Brugmansia suaveolens</i> (Humb. & Bonpl. Ex Willd.) Sweet (1818)
Zamioculca (Or)	Araceae	<i>Zamioculcas zamiifolia</i> (Lodd.) Engl. (1905)

Or: ornamental; M: medicinal; A: arboreal; S: shrub; W: weed.

Table 2. Classificatory ranking of extract effect under time influence expressed in percentage.

Plant Specie	TIME (hours)										General Average
	0	C or S (%)	6	C or S (%)	12	C or S (%)	24	C or S (%)	30	C or S (%)	
<i>Agave angustifolia</i>	8.583 Aa	-14.2	8.583 Aa	-14.17	9.041 Aa	-9.59	13.667 Bc	36.67	14.500 Bc	45	10.875
<i>Aloe vera</i>	9.833 Aa	-1.67	10.667 Aa	6.67	14.208 Bb	42.08	23.708 Cf	137.08	22.792 Cf	127.92	16.242
<i>Garcinia gardneriana</i>	8.917 Aa	-10.8	8.916 Aa	-10.84	10.083 Aa	0.83	10.458 Aa	4.58	11.167 Ab	11.67	9.908
<i>Costus spicatus</i>	8.708 Aa	-12.9	10.000 Aa	0	11.750 Ba	17.5	16.667 Cd	66.67	18.208 Cd	82.08	13.067
<i>Baccharis trimera</i>	10.000 Aa	0	10.083 Aa	0.83	10.167 Aa	1.67	15.125 Bc	51.25	24.625 Cg	146.25	14.000
<i>Cedreia fissilis</i>	9.500 Aa	-5	9.500 Aa	-5	9.625 Aa	-3.75	9.708 Aa	-2.92	9.542 Aa	-4.58	9.575
<i>Schefflera arboricola</i>	10.000 Aa	0	11.000 Aa	10	13.250 Bb	32.5	20.875 Ce	108.75	25.333 Dg	153.33	16.092
<i>Schinus molle</i>	10.208 Aa	2.08	14.667 Bc	46.67	17.000 Cc	70	21.000 De	110	18.583 Cd	85.83	16.292
<i>Clusia variegata</i>	9.958 Aa	-0.42	10.917 Aa	9.17	12.375 Ab	23.75	18.583 Bd	85.83	19.583 Bd	95.83	14.283
<i>Dieffenbachia seguine</i>	10.208 Aa	2.08	10.208 Aa	2.08	10.333 Aa	3.33	13.917 Bc	39.17	15.792 Bc	57.92	12.092
<i>Euphorbia milii</i>	9.292 Aa	-7.08	9.292 Aa	-7.08	9.458 Aa	-5.42	10.292 Aa	2.92	11.375 Ab	13.75	9.942
<i>Cecropia pachystachya</i>	8.708 Aa	-12.9	8.708 Aa	-12.92	10.125 Aa	1.25	15.875 Bc	58.75	16.625 Bc	66.25	12.008
<i>Psychotria carthagenensis</i>	9.208 Aa	-7.92	9.333 Aa	-6.67	9.417 Aa	-5.83	9.333 Aa	-6.67	8.917 Aa	-10.83	9.242
<i>Sansevieria trifasciata</i>	10.375 Aa	3.75	10.958 Aa	9.58	11.083 Aa	10.83	11.292 Ab	12.92	11.333 Ab	13.33	11.008
<i>Ficus carica</i>	10.958 Aa	9.58	12.625 Ab	26.25	12.833 Ab	28.33	11.208 Ab	12.08	10.667 Ab	6.67	11.658
<i>Peschiera fuchsiaefolia</i>	10.042 Aa	0.42	10.042 Aa	0.42	10.208 Aa	2.08	9.292 Aa	-7.08	8.292 Aa	-17.08	9.575
<i>Petiveria alliaceae</i>	9.667 Aa	-3.33	9.667 Aa	-3.33	10.875 Aa	8.75	10.333 Aa	3.33	10.000 Aa	0	10.108
<i>Mentha villosa</i>	9.667 Aa	-3.33	10.750 Aa	7.5	17.250 Bc	72.5	25.542 Cf	155.42	27.042 Ch	170.42	18.050
<i>Genipa americana</i>	9.417 Aa	-5.83	9.542 Aa	-4.58	9.792 Aa	-2.08	10.042 Aa	0.42	9.958 Aa	-0.42	9.750
<i>Ligustrum lucidum</i>	9.833 Aa	-1.67	9.792 Aa	-2.08	9.958 Aa	-0.42	9.708 Aa	-2.92	10.125 Aa	1.25	9.883
<i>Artemisia absinthium</i>	9.375 Aa	-6.25	9.375 Aa	-6.25	9.625 Aa	-3.75	9.708 Aa	-2.92	9.792 Aa	-2.08	9.575
<i>Carica papaya</i>	10.416 Aa	4.16	9.250 Aa	-7.5	10.708 Aa	7.08	9.625 Aa	-3.75	8.500 Aa	-15	9.700
<i>Manihot esculenta</i>	10.375 Aa	3.75	12.000 Ab	20	12.625 Bb	26.25	13.458 Bc	34.58	14.208 Bc	42.08	12.533
<i>Solanum americanum</i>	11.000 Aa	10	11.000 Aa	10	11.625 Aa	16.25	11.417 Ab	14.17	10.000 Aa	0	11.008
<i>Erythrina verna</i>	9.333 Aa	-6.67	9.333 Aa	-6.67	10.583 Aa	5.83	14.000 Bc	40	21.125 Ce	111.25	12.875
<i>Morinda citrifolia</i>	10.667 Aa	6.67	12.500 Bb	25	12.708 Bb	27.08	11.708 Bb	17.08	10.167 Aa	1.67	11.550
<i>Gallesia integrifolia</i>	10.000 Aa	0	10.667 Aa	6.67	11.292 Aa	12.92	11.833 Ab	18.33	12.333 Ab	23.33	11.225

Continue...

Table 2. Continuation.

Plant Specie	TIME (hours)										General Average
	0	C or S (%)	6	C or S (%)	12	C or S (%)	24	C or S (%)	30	C or S (%)	
<i>Bauhinia forficata</i>	10.208 Aa	2.08	10.167 Aa	1.67	10.458 Aa	4.58	10.583 Aa	5.83	10.542 Aa	5.42	10.392
<i>Jatropha gossypifolia</i>	9.750 Aa	-2.5	9.750 Aa	-2.5	9.875 Aa	-1.25	9.333 Aa	-6.67	9.167 Aa	-8.33	9.575
<i>Mentha</i> sect. <i>pulegium</i>	9.625 Aa	-3.75	9.542 Aa	-4.58	10.167 Aa	1.67	14.333 Bc	43.33	16.000 Bc	60	11.933
<i>Sambucus nigra</i>	9.833 Aa	-1.67	9.958 Aa	-0.42	10.583 Aa	5.83	12.208 Bb	22.08	12.208 Bb	22.08	10.958
<i>Brugmansia suaveolens</i>	9.125 Aa	-8.75	9.125 Aa	-8.75	9.792 Aa	-2.08	10.958 Ab	9.58	15.417 Bc	54.17	10.883
<i>Zamioculcas zamiifolia</i>	9.583 Aa	-4.17	9.750 Aa	-2.5	11.125 Ba	11.25	11.547 Bb	15.47	12.250 Bb	22.5	10.851
C.V. (%)	30.84										
Maximum		10		46.67		72.5		155.42		170.42	18.0502
Minimum		-14.2		-14.17		-9.59		-7.08		-17.08	9.2416

C: controller extract; S: stimulator extract. Means followed by the same lowercase letter in column and by the same capital letter in line did not differ significantly from each other with the Scott-Knott test, at 5% error probability.

Solanum americanum, *Morinda citrifolia*, *Gallsia integrifolia*, *Sambucus nigra*, and *Brugmansia suaveolens*. All others presented some variation at some point.

Extract concentration had more influence (Table 3) on control percentages than time (Table 2). Most lethal yields were caused by *C. papaya* (-66% at 20%; -33.7% at 10%), *Euphorbia milii* (-37% at 20%), *P. carthagenensis* (-25.5% at 2.5%), *Clusia variegata* (-22% at 20%), *Zamioculcas zamiifolia* (-21.5% at 20%), *P. fuchsiaeifolia* (-20.5% at 10%), and *Cedreia fissilis* (-19.5% at 20%). Other extracts displaying control were *Ligustrum lucidum* (-18.5% at 1.25%), *Artemisia absinthium* (-18.5% at 2.5%), *C. fissilis* (-17.5% at 1.25%), *Garcinia gardneriana* (-17% at 1.25%), *Jatropha gossypifolia* (-17% at 2.5%; -16% at 20%), and *Petiveria alliaceae* (-14% at 20%). Others presented lower control indexes.

On the other hand, several extracts stimulated nematode reproduction, an undesirable reaction. In fact, most results varied from a slight to a huge reproduction increase, resulting in many juveniles. There were 24 extracts surpassing 50% on stimuli. *M. villosa* only stimulated nematodes, being higher at concentrations of 10% (148%), and 2.5% (131.5%), followed by *Aloe vera* (123% at 5%), *Schinus molle* (112.5% at 10%), *Schefflera arboricola* (93.5% at 5%), *C. variegata* (89% at 5%), and *S. molle* (88% at 5%). Concentration influenced extract effect, especially for *C. variegata*, that also had controlled nematodes (-22% at 20%).

Surprisingly, among the plants randomly chosen, some ornamentals, such as *C. variegata*, *A. angustifolia*, *E. milii*, *Z. zamiifolia*, *Dieffenbachia seguine*, and *Sansevieria trifasciata*

killed nematodes in a specific concentration, especially at 20%, although they were stimulated in others. *E. milii* kept population low and stable in concentrations lower than 20%. In the ornamental group, only *B. suaveolens* and *S. arboricola* did not control nematodes at any concentration (Table 3). *P. carthagenensis* controlled nematodes in all concentrations, but higher concentrations did not determine higher control percentages, revealing no linear relation between dose and its respective controlling percentage (Table 4).

In respect to regression analysis, ANOVA showed significance for time, dose and their interaction (data not presented). As a biological characteristic, extracts did not express linear behavior, fitting better at second degree polynomial equation (Table 4). Graphs allow a better individual interpretation of time, and dose influences contrasting extracts. There was an additional effect for *M. villosa* (Fig. 1A), where longer exposure time increased reproduction, as well as dose, although dose was not significant (Table 4). Doses of *C. papaya* influenced nematodes more as to death, whereas time only kept population stable (Fig. 1B). For *Z. zamiifolia*, nematodes were not stimulated for time or dose (Fig. 2A); on the other hand, time caused nematode reproduction for *C. pachystachya*, whereas dose had no significant effects on it (Fig. 2B).

After evaluating the effect of six concentrations, the balance of alive nematodes was influenced by this combination five times more, and both Scott-Knott and regression tests masked some remarkable results, such as for *C. papaya*, that controlled 100% of nematodes after 24 hours at 10% w/v, and after 12 hours at 20% w/v.

Table 3. Classificatory ranking of extract reaction under dose influence expressed in percentage of effect.

Plant Specie	CONCENTRATION (%)										General Average
	1,25	C or S (%)	2,5	C or S (%)	5	C or S (%)	10	C or S (%)	20	C or S (%)	
<i>Agave angustifolia</i>	13.200 Bc	32	9.750 Aa	-2,5	11.650 Bb	16,5	11.150 Bc	11,5	9.000 Ac	-10	11,15
<i>Aloe vera</i>	14.650 Ac	46,5	17.250 Bd	72,5	22.300 Cf	123	14.400 Ac	44	18.350 Bf	83,5	17,25
<i>Garcinia gardneriana</i>	8.300 Aa	-17	9.850 Aa	-1,5	10.100 Aa	1	9.650 Ab	-3,5	11.050 Ad	10,5	9,85
<i>Costus spicatus</i>	12.750 Ab	27,5	16.450 Bd	64,5	11.750 Ab	17,5	14.750 Bf	47,5	11.950 Ae	19,5	12,75
<i>Baccharis trimera</i>	17.300 Bd	73	14.300 Ac	43	14.650 Ac	46,5	14.150 Ac	41,5	13.100 Ae	31	14,30
<i>Cedreria fissilis</i>	10.950 Bb	9,5	10.500 Bb	5	9.200 Aa	-8	8.250 Aa	-17,5	8.050 Ac	-19,5	9,20
<i>Schefflera arboricola</i>	17.750 Bd	77,5	17.500 Bd	75	19.350 Be	93,5	17.150 Be	71,5	14.300 Ae	43	17,50
<i>Schinus molle</i>	17.000 Bd	70	14.750 Ac	47,5	18.800 Be	88	21.250 Cf	112,5	15.450 Af	54,5	17,00
<i>Clusia variegata</i>	15.850 Bd	58,5	15.900 Bd	59	18.900 Ce	89	16.750 Be	67,5	7.800 Ac	-22	15,90
<i>Dieffenbachia seguine</i>	10.750 Ab	7,5	13.700 Bc	37	13.400 Bc	34	9.300 Ab	-7	14.900 Bf	49	13,40
<i>Euphorbia milii</i>	10.400 Bb	4	9.250 Ba	-7,5	12.600 Bb	26	10.600 Bb	6	6.300 Ab	-37	10,40
<i>Cecropia pachystachya</i>	11.750 Ba	17,5	13.850 Ac	38,5	11.400 Ab	14	12.550 Ac	25,5	12.000 Ae	20	12,00
<i>Psychotria carthagenensis</i>	8.900 Aa	-11	7.450 Aa	-25,5	9.850 Aa	-1,5	9.800 Ab	-2	8.950 Ac	-10,5	8,95
<i>Sansevieria trifasciata</i>	9.150 Aa	-8,5	12.050 Bb	20,5	10.200 Aa	2	10.350 Ab	3,5	13.800 Be	38	10,35
<i>Ficus carica</i>	14.050 Bc	40,5	11.250 Ab	12,5	11.350 Ab	13,5	12.750 Bc	27,5	10.050 Ad	0,5	11,35
<i>Peschiera fuchsiaefolia</i>	8.850 Aa	-11,5	10.250 Aa	2,5	10.500 Aa	5	7.950 Aa	-20,5	9.400 Ac	-6	9,40
<i>Petiveria alliaceae</i>	13.350 Bc	33,5	9.400 Aa	-6	8.950 Aa	-10,5	9.850 Ab	-1,5	8.600 Ac	-14	9,40
<i>Mentha villosa</i>	16.350 Ad	63,5	23.150 Be	131,5	16.000 Ad	60	24.800 Bg	148	17.500 Af	75	17,50
<i>Genipa americana</i>	9.300 Aa	-7	11.100 Ab	11	9.150 Aa	-8,5	9.550 Ab	-4,5	8.900 Ac	-11	9,30
<i>Ligustrum lucidum</i>	8.150 Aa	-18,5	9.800 Aa	-2	10.100 Aa	1	10.700 Ab	7	10.050 Ad	0,5	10,05
<i>Artemisia absinthium</i>	8.950 Aa	-10,5	8.150 Aa	-18,5	11.400 Bb	14	9.800 Bb	-2	8.650 Ac	-13,5	8,95
<i>Carica papaya</i>	14.900 Dc	49	11.300 Cb	13	11.850 Cb	18,5	6.250 Ba	-37,5	3.400 Aa	-66	11,30
<i>Manihot esculenta</i>	12.100 Ab	21	11.600 Ab	16	10.400 Aa	4	14.750 Bf	47,5	15.850 Bf	58,5	12,10
<i>Solanum americanum</i>	11.400 Ab	14	10.650 Ab	6,5	11.350 Ab	13,5	9.700 Ab	-3	12.450 Ae	24,5	11,35
<i>Erythrina verna</i>	14.700 Bc	47	17.750 Cd	77,5	13.300 Bc	33	10.450 Ab	4,5	10.550 Ad	5,5	13,30
<i>Morinda citrifolia</i>	12.900 Ab	29	12.300 Ab	23	9.950 Aa	-0,5	11.800 Ac	18	11.850 Ae	18,5	11,85
<i>Gallesia integrifolia</i>	12.200 Ab	22	12.300 Ab	23	11.300 Ab	13	9.950 Ab	-0,5	11.100 Ae	11	11,30

Continue...

Table 3. Continuation.

Plant Specie	CONCENTRATION (%)										General Average
	1,25	C or S (%)	2,5	C or S (%)	5	C or S (%)	10	C or S (%)	20	C or S (%)	
<i>Bauhinia forficata</i>	10.600 Ab	6	10.950 Ac	9,5	8.750 Aa	-12,5	10.600 Ab	6	10.950 Ad	9,5	10,60
<i>Jatropha gossypifolia</i>	11.800 Bb	18	8.300 Aa	-17	9.000 Aa	-10	9.450 Ab	-5,5	8.400 Ac	-16	9,00
<i>Mentha</i> sect. <i>pulegium</i>	11.100 Ab	11	9.950 Aa	-0,5	11.750 Ab	17,5	13.450 Bc	34,5	14.850 Bf	48,5	11,75
<i>Sambucus nigra</i>	11.750 Ab	17,5	11.350 Ab	13,5	12.250 Ab	22,5	10.450 Ab	4,5	9.450 Ac	-5,5	11,35
<i>Brugmansia suaveolens</i>	11.950 Ab	19,5	10.500 Ab	5	10.700 Aa	7	11.550 Ac	15,5	10.100 Ad	1	10,70
<i>Zamioculcas zamiifolia</i>	12.200 Cb	22	10.150 Ba	1,5	10.300 Ba	3	14.100 Cd	41	7.850 Ac	-21,5	10,30
C.V. (%)	30,84										
Maximum	77,5		131,5		123		148		83,5		17,5
Minimum	-18,5		-25,5		-12,5		-37,5		-66		8,95

C: controller extract; S: stimulator extract. Means followed by the same lowercase letter in column and by the same capital letter in line did not differ significantly from each other with the Scott-Knott test, at 5% error probability.

All extracts showed significance to the combination between time and dose, although some of them presented no influence by dose or time variable by itself. For the extracts in which nematodes had no influence of the variables, population was kept stable throughout the experiment (Table 4).

DISCUSSION

The search for new plants with anthelmintic potential may be random or focused on no host species to phytonematodes, since these plants could contain nematicide compounds (MARTINS; SANTOS, 2016). Seen that, WIRATNO et al. (2009) tested nematicidal activity of 17 plant extracts against *Meloidogyne incognita* with an ethanolic extraction of chemical compounds, composing buds, leaves, flowers, roots, seeds, and stems. According to them, tobacco, clove and betelvine presented high toxicity levels, killing over 80% of nematodes at a 5 mg mL⁻¹ dosage, followed by sweet flag, pyrethrum, and citronella, whose control potential ranged from 10 to 20%. In our study, no extract was more effective than 66%. One explanation lies on the fact that extraction method changes effectiveness of secondary metabolites against phytopathogens, also related to the influence of temperature (VENTUROSOSO et al., 2010).

KLIMPEL et al. (2011) evaluated the effectiveness of 13 plant extracts submitted to different extraction procedures (aqueous, ethanolic, methanolic, or chloroform) against three nematodes species. According to them, each methodology eluted compounds in different grades, presenting a varied

potential. Aqueous extracts performed better control *in vitro*, especially after 24 hours.

Given that, aqueous extract of *Ficus carica* allowed nematodes reproduction at any dosage and time (Tables 2 and 3), whereas its methanolic extraction and use against *P. redivivus* showed inconsistent findings, ranging from less than 1% (CUNHA et al., 2003) to 96.2% (5 mg/mL) after 72 hours (LIU et al., 2011).

HONG et al. (2007) tested 30 plants with ethanolic extraction to set their control potential against *Bursaphelenchus xylophilus* and *P. redivivus*. Among them, *Sapiumse biserum*, *Nerium indicum*, *Magnolia grandiflora*, *Michelia hedyosperma*, *Zingiber striolatum*, *Punica granatum*, and *Edgeworthia chrysantha* killed over 40% nematodes within 24 hours. In this study, only *C. papaya* overcame this mortality percentage, whose anthelmintic effect is well known in literature, although its methanolic extract killed only 15.36% of the initial population of *P. redivivus* (20-30 individuals) after 48 hours (CUNHA et al., 2003).

Extraction methods seem to vary among plant species, once methanolic extracts prepared from 24 plants were used to verify their nematicide potential against *P. redivivus*, revealing a reduction of over 94% by *Leucaena leucocephala* and *Paspalum notatum* (CUNHA et al., 2003), whereas other six plant extracts varied their efficiency from 11.054 to 36.33% after 48 hours. These control indexes surpass most of ours, possibly because authors applied the Tukey test, that compares all possible treatment medias among themselves, two-by-two, and the Scott-Knott test groups treatments minimize variation within groups and maximize variation among groups to

compare them without ambiguity, leading to a lesser groups number formation.

It shall be said that statistical analysis also plays an important role. The Scott-Knott test is ideal to compare a large number of treatments for grouping related media without ambiguity (BHERING et al., 2008), considering that other tests may present data overlap (CANTERI et al., 2001). Nonetheless, results were hidden by multiple comparison of media (Tables 2 and 3), because statistical breakdown evaluates general dose

at each time (vice-versa), and, evidently, any concentration in earlier evaluation periods had less effects. This is why, statistically, the highest control did not exceed 60% when it actually did happen, especially for *C. papaya* extract, that killed 100% of nematodes at 20% w/v after 12, 24 and 30 hours.

Mortality rates increased during exposure time (ELBADRI et al., 2008). However, only few extracts fit in such pattern, especially *C. papaya*, *P. fuchsiaefolia*, and *P. carthagenensis* (Table 2).

Table 4. Multiple linear regression test results.

Specie	Equation	R ²
<i>Carica papaya</i>	$y = 13.543^{**} - 0.493 x^{**} - 0.046 x^{2ns}$	42.31
<i>Agave angustifolia</i>	$y = 8.323^{**} - 0.109 x^{ns} + 0.226 x^{2**}$	59.80
<i>Aloe vera</i>	$y = 7.602^{**} + 0.193 x^{ns} + 0.513 x^{2**}$	58.69
<i>Garcinia gardneriana</i>	$y = 8.387^{**} + 0.065 x^{**} + 0.077 x^{2**}$	57.45
<i>Costus spicatus</i>	$y = 8.300^{**} - 0.0054 x^{ns} + 0.333 x^{2**}$	65.53
<i>Baccharis trimera</i>	$y = 7.767^{**} - 0.030 x^{ns} + 0.446 x^{2**}$	44.91
<i>Cedreia fissilis</i>	$y = 10.471^{**} - 0.147 x^{**} + 0.004 x^{2ns}$	69.97
<i>Schefflera arboricola</i>	$y = 8.555^{**} - 0.012 x^{ns} + 0.529 x^{2**}$	65.20
<i>Schinus molle</i>	$y = 11.127^{**} + 0.150 x^{ns} + 0.291 x^{2**}$	35.80
<i>Clusia variegata</i>	$y = 10.923^{**} - 0.269 x^{ns} + 0.354 x^{2**}$	39.85
<i>Dieffenbachia seguine</i>	$y = 8.396^{**} + 0.131 x^{*} + 0.198 x^{2**}$	54.75
<i>Euphorbia milii</i>	$y = 10.209^{**} - 0.191 x^{**} + 0.067 x^{2ns}$	33.87
<i>Cecropia pachystachya</i>	$y = 7.467^{**} + 0.023 x^{ns} + 0.305 x^{2**}$	66.61
<i>Psychotria carthagenensis</i>	$y = 9.404^{**} - 0.007 x^{ns} - 0.008 x^{2ns}$	0.81
<i>Sansevieria trifasciata</i>	$y = 9.570^{**} + 0.161 x^{**} + 0.028 x^{2ns}$	53.38
<i>Ficus carica</i>	$y = 12.614^{**} - 0.074 x^{ns} - 0.033 x^{2ns}$	9.03
<i>Peschiera fuchsiaefolia</i>	$y = 10.174^{**} - 0.051 x^{ns} - 0.056 x^{2*}$	18.85
<i>Petiveria alliacea</i>	$y = 10.730^{**} - 0.130 x^{*} + 0.016 x^{2ns}$	16.98
<i>Mentha villosa</i>	$y = 7.537^{**} + 0.201 x^{ns} + 0.640 x^{2**}$	59.11
<i>Genipa americana</i>	$y = 9.898^{**} - 0.068 x^{**} + 0.020 x^{2ns}$	35.48
<i>Ligustrum lucidum</i>	$y = 9.571^{**} + 0.036 x^{ns} + 0.005 x^{2ns}$	7.12
<i>Artemisia absinthium</i>	$y = 9.625^{**} - 0.040 x^{ns} + 0.015 x^{2ns}$	6.99
<i>Manihot esculenta</i>	$y = 9.184^{**} + 0.265 x^{**} + 0.114 x^{2**}$	62.06
<i>Solanum americanum</i>	$y = 10.923^{**} + 0.060 x^{ns} - 0.021 x^{2ns}$	12.14
<i>Erythrina verna</i>	$y = 8.863^{**} - 0.192 x^{ns} + 0.364 x^{2**}$	46.73
<i>Morinda citrifolia</i>	$y = 11.878^{**} + 0.014 x^{ns} - 0.029 x^{2ns}$	3.32
<i>Bauhinia forficata</i>	$y = 10.040^{**} + 0.023 x^{ns} + 0.014 x^{2ns}$	7.12
<i>Gallesia integrifolia</i>	$y = 10.422^{**} - 0.040 x^{ns} + 0.073 x^{2**}$	42.81
<i>Jatropha gossypifolia</i>	$y = 10.527^{**} - 0.099 x^{*} - 0.022 x^{2ns}$	22.95
<i>Mentha sect. pulegium</i>	$y = 7.061^{**} + 0.236 x^{**} + 0.233 x^{2**}$	76.87
<i>Sambucus nigra</i>	$y = 10.246^{**} - 0.095 x^{**} + 0.092 x^{2**}$	64.30
<i>Brugmansia suaveolens</i>	$y = 8.447^{**} - 0.034 x^{ns} + 0.184 x^{2**}$	42.73
<i>Zamioculcas zamiifolia</i>	$y = 10.257^{**} - 0.107 x^{*} + 0.089 x^{2**}$	29.98

R²: coefficient of determination; **significant with $p \leq 0.01$; *significant with $p \leq 0.05$, ns: non-significant ($p > 0.05$); y: alive nematodes estimative; x: dose independent variable; x²: time independent variable.

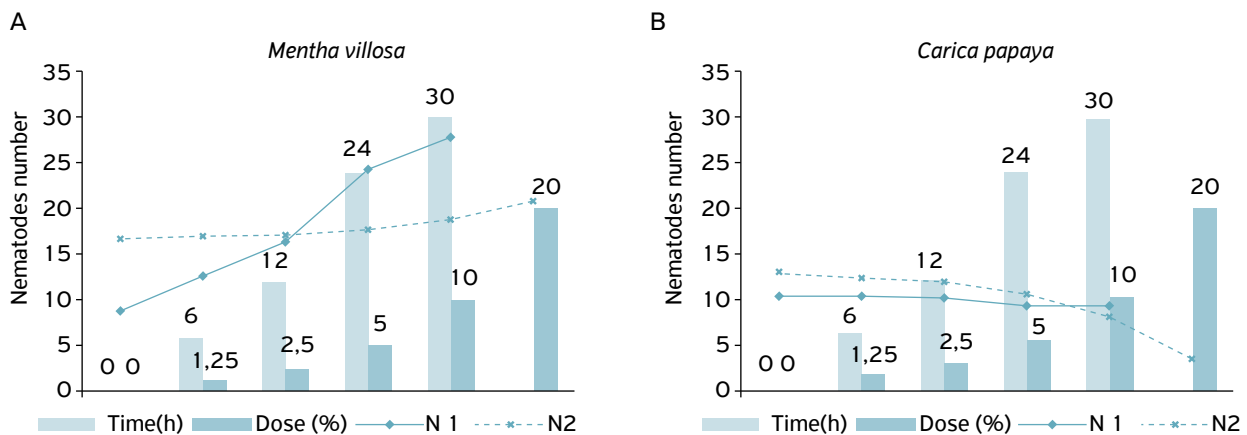
As to ornamental plants, CUNHA et al. (2003) did not notice significant effects on *P. redivivus* control after 48 hours by methanolic extracts of *Asparagus densiflorus*, *Pelargonium hortorum*, *Zinnia elegans*, *Tagetes erecta*, and *Euphorbia pulcherrima*, which belongs to the Euphorbiaceae family, just like *E. milii* — that kept population low and stable until 12 hours (Table 2). However, the authors reported control set by *Dendranthema grandiflorum* (36.33%), and *Bougainvillea glabra* (23.39 %). In addition, KLIMPEL et al. (2011) did not observe any immediate effect (0 hours) of aqueous extracts on nematodes movement, exactly like what we found.

Most authors do not consider nematodes reproduction on research, solely counting the killed ones. Despite that, we considered this an important factor, because some plant extracts may stimulate nematodes, as noticed by many species in different concentrations and times (Tables 1 and 2). Some of these extracts had killed some individuals, but stimulated

them more, and the final balance of alive nematodes masked those that were dead.

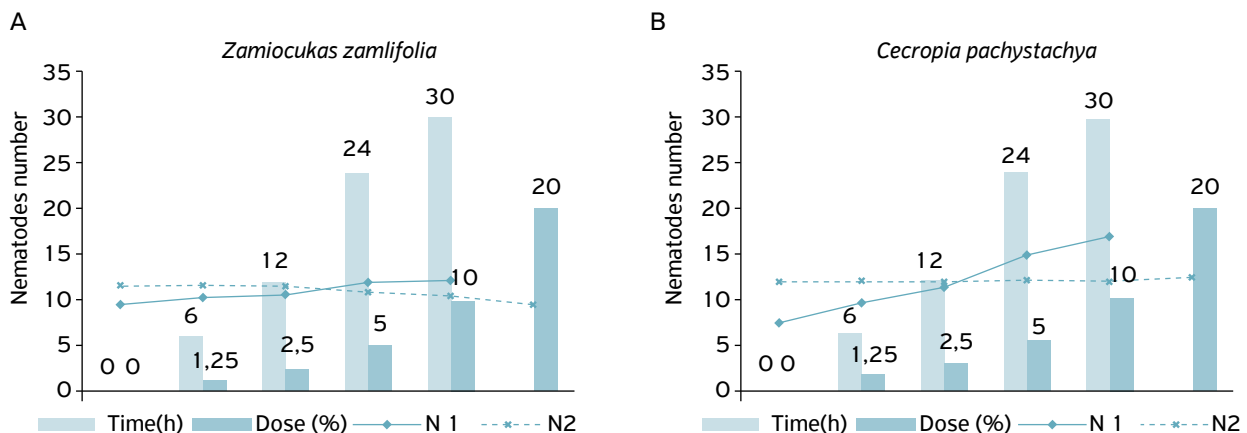
ELBADRI et al. (2008) tested methanol and hexane extracts (0.05% concentration) of 21 medicinal plants for their toxicity against *M. incognita*. Nematodes were sensitive to all of them at some level, mainly after 72 hours. In this study, all concentrations of *Baccharis trimera*, *M. villosa*, and *A. vera* led to an increase of over than 30% in nematode reproduction (Table 3). *Petiveria alliaceae* had controlled over 10% of nematodes at 5 and 20% w/v concentrations, whereas *A. absinthium* did not control only at 5% w/v, and *M. sect. pulegium* had irrelevant control (0.5%) at 2.25% w/v.

MARTINS; SANTOS (2016) prepared aqueous extracts of 10 medicinal plants to confront with *M. incognita* race 2 for 48 hours, counting inactive nematodes. Juveniles were put into water for 24 hours to analyze any eventual recovery. According to them, *Eclipta alba*, *Ocimum basilicum*, *Artemisia vulgaris*, *Justicia pectoralis* var. *stenophylla*, *Spigelia*



N1: alive nematodes influenced by time; N2: alive nematodes influenced by dose.

Figure 1. *Mentha villosa* (A) and *Carica papaya* (B) extracts, respectively.



N1: alive nematodes influenced by time; N2: alive nematodes influenced by dose.

Figure 2. *Zamioculcas zamiifolia* (A) and *Cecropia pachystachya* (B) extracts, respectively.

anthelmia, and *Chenopodium ambrosioides* killed over 75% of nematodes at the concentration of 10%, prepared by either infusion or maceration. Nonetheless, *M. x villosa* was the less effective extract against *M. incognita* race 2, and a stimulator to *P. redivivus* at any concentration (Table 3).

Panagrellus redivivus as a free-living nematode moves freely in water; the same behavior was seen when nematodes were exposed to many extracts, especially the stimulator ones. This way, it was easy to check dead individuals, because their body got very strict or even end-curved, without any physical moves. WIRATNO et al. (2009) reported shape changings, depending on the plant extract. An excessive movement for stimulator extracts was observed, to which the presence of many juveniles was very common, but it did not provoke death, unlikely noticed by WIRATNO et al. (2009). MARTINS; SANTOS (2016) demonstrated differences on nematicide and static effects.

ELBADRI et al. (2008) assumed that all extracts from seed had a higher nematicidal activity, matching with those we found (*C. papaya*). Seeds present higher chemical concentration of many soluble substances, such as alkaloids, tannin, phenolic compounds, and others (MARCOS FILHO, 2005).

In vitro evaluations aim at ranking extract plants to screen the best ones for further tests. MATEUS et al. (2014) evaluated the effect of biweekly application of *Erythrina mulungu* Mart. ex Benth aqueous extract, observing a reduction of 40% in *M. incognita* infectiveness in tomato roots, and 97.1% less eggs. This activity against nematodes is believed to result from secondary metabolites. Our *in vitro* results revealed the time factor, increasing *P. redivivus* population when treated with *Erythrina verna* (Table 4).

FERREIRA et al. (2013) tried aqueous extracts of *Sphagneticola trilobata*, *Tagetes patula*, *Tithonia diversifolia*, *Tridax procumbens*, *Unxia suffruticosa*, and *Zinnia peruviana* on *M. incognita*, recording eggs outbreak over 85% *in vitro*; when they were tested in tomato plants, no statistical difference for root weight and nematode reproduction index was noticed, compared to treatment with water (control). Therefore, our results are promising, although they may not display the same reaction in *in vivo* situations (KLIMPEL et al., 2011), demanding future assays.

In summary, *P. redivivus* was better controlled by *Carica papaya*, *Euphorbia milii*, *Psychotria carthagenensis*, *Clusia variegata*, *Zamioculcas zamiifolia*, *Peschiera fuchsiaeifolia*, and *Cedreia fissilis* aqueous extracts, and highly stimulated by *Mentha villosa*, *Aloe vera*, *Schinus molle*, and *Schefflera arboricola*.

The hypothesis was confirmed, because some ornamental plants randomly chosen showed a control effect on *P. redivivus*, indicating the existence of new nematicides compounds. We first reported the action of *C. variegata*, *Z. zamiifolia*, and *P. carthagenensis* on *P. redivivus*.

For some extracts, dose and time had a concomitant influence on nematodes control, whereas for others, only one of such parameters was significant.

ACKNOWLEDGEMENTS

Universidade Federal do Paraná – Setor Palotina supported research.

REFERENCES

- BHERING, L.L.; CRUZ, C.D.; VASCONCELOS, E.S.; FERREIRA, A.; RESENDE JUNIOR, M.F.R. Alternative methodology for Scott-Knott test. *Crop Breeding and Applied Biotechnology*, v.8, n.1, p.9-16, 2008. <http://dx.doi.org/10.12702/1984-7033.v08n01a02>
- CAMPOS, H.D.; CAMPOS, V.P.; SILVA, J.R.C.; SILVA, L.H.C.P.; COSTA, L.S.A.S.C.; TERRA, W.C. Atração e penetração de *Meloidogyne javanica* e *Heterodera glycines* em raízes excisadas de soja. *Ciência Rural*, v.41, n.9, p.1496-1502, 2011. <http://dx.doi.org/10.1590/S0103-84782011000900002>
- CANTERI, M.G.; ALTHAUS, R.A.; VIRGENS FILHO, J.S.; GIGLIOTI, E.A.; GODÓI, C.V. Sasm-Agri-Sistema para análise e separação de médias em experimentos agrícolas pelos métodos de Scott-Knott, Tukey e Duncan. *Revista Brasileira de Agrocomputação*, v.1, n.2, p.18-24, 2001.
- CUNHA, F.R.; OLIVEIRA, D.F.; CAMPOS, V.P. Extratos vegetais com propriedades nematicidas e purificação do princípio ativo do extrato de *Leucaena leucocephala*. *Fitopatologia Brasileira*, v.28, n.4, p.438-441, 2003. <http://dx.doi.org/10.1590/S0100-41582003000400017>
- DE ALMEIDA ENGLER J., FAVERY, B. The plant cytoskeleton remodelling in nematode induced feeding sites. In: JONES, J.; GHEYSEN, G.; FENOLL, C. *Genomics and Molecular Genetics of Plant-Nematode Interactions*. Dordrecht: Springer, 2011. p.369-393. https://doi.org/10.1007/978-94-007-0434-3_18
- DIAS, M.H.; BARBOSA, J.A.; PETERS, F.F.; STANGARLIN, J.R.; ESTEVES, R.L. Controle alternativo de *Meloidogyne incognita* em tomateiro. *Scientia Agraria Paranaensis*, v.15, n.4 p.421-426, 2016. <http://dx.doi.org/10.1818/sap.v15i4.12491>

- ELBADRI, G.A.; LEE, D.W.; PARK, J.C.; YU, H.B.; CHOO, H.Y. Evaluation of various plant extracts for their nematicidal efficacies against juveniles of *Meloidogyne incognita*. *Journal of Asia-Pacific Entomology*, v.11, n.2, p.99-102, 2008. <https://doi.org/10.1016/j.aspen.2008.04.004>
- FERREIRA, D. F. Sisvar: um sistema computacional de análise estatística. *Ciência e Agrotecnologia*, v.35, n.6, p.1039-1042, 2011. <http://dx.doi.org/10.1590/S1413-70542011000600001>
- FERREIRA, I.C.M.; SILVA, G.S.; NASCIMENTO, F.S. Efeito de extratos aquosos de espécies de Asteraceae sobre *Meloidogyne incognita*. *Summa Phytopathologica*, v.39, n.1, p.40-44, 2013. <http://dx.doi.org/10.1590/S0100-54052013000100007>
- GARDIANO, C.G.; FERRAZ, S.; LOPES, E.A.; FERREIRA, P.A.; AMORA, D.X.; FREITAS, L.G. Evaluation of plant aqueous extracts, added into the soil on *Meloidogyne javanica* (Treub, 1885) Chitwood, 1949. *Ciências Agrárias*, v.30, n.3, p.551-556, 2009. <http://dx.doi.org/10.5433/1679-0359.2009v30n3p551>
- HONG, L.; LI, G.; ZHOU, W.; WANG, X.; ZHANG, K. Screening and isolation of a nematicidal sesquiterpene from *Magnolia grandiflora* L. *Pest Management Science*, v.63, n.3, p.301-305, 2007. <https://doi.org/10.1002/ps.1337>
- ISMAN, M.B. Plant essential oils for pest and disease management. *Crop Protection*, v.19, n.8-10, p.603-608, 2000. [https://doi.org/10.1016/S0261-2194\(00\)00079-X](https://doi.org/10.1016/S0261-2194(00)00079-X)
- JONES, J.T.; HAEGEMAN, A.; DANCHIN, E.G.J.; GAUR, H.S.; HELDER, J.; JONES, M.G.K.; KIKUCHI, T.; MANZANILLA-LÓPEZ, R.; PALOMARES-RIUS, J.E.; WESEMAEL, W.I.M.M.L.; PERRY, R.N. Top 10 plant-parasitic nematodes in molecular plant pathology. *Molecular Plant Pathology*, v.14, n.9, p.946-961, 2013. <https://doi.org/10.1111/mpp.12057>
- KLIMPEL, S.; ABDEL-GHAFFAR, F.; AL-RASHEID, K. A. S.; AKSU, G.; FISCHER, K.; STRASSEN, B.; MEHLHORN, H. The effects of different plant extracts on nematodes. *Parasitology Research*, v.108, n.4, p.1047-1054, 2011. <https://doi.org/10.1007/s00436-010-2168-4>
- LIU, F.; YANG, Z.; ZHENG, X.; LUO, S.; ZHANG, K.; LI, G. Nematicidal coumarin from *Ficus carica* L. *Journal of Asia-Pacific Entomology*, v.14, n.1, p.79-81, 2011. <https://doi.org/10.1016/j.aspen.2010.10.006>
- MARCOS FILHO, J. Composição química de sementes (reservas armazenadas). In: MARCOS FILHO, J. *Fisiologia de sementes de plantas cultivadas*. Piracicaba: Fealq, 2005. 495p.
- MARTINS, M.C.B.; SANTOS, C.D.G. Ação de extratos de plantas medicinais sobre juvenis de *Meloidogyne incognita* raça 2. *Revista Ciência Agronômica*, v.47, n.1, p.135-142, 2016. <https://doi.org/10.5935/1806-6690.20160016>
- MATEUS, M.A.F.; FARIA, C.M.D.R.; BOTELHO, R.V.; DALLEMOLE-GIARETTA, R.; FERREIRA, S.G.M.; ZALUSKI, W.L. Extratos aquosos de plantas medicinais no controle de *Meloidogyne incognita* (Kofoid e White, 1919) Chitwood, 1949. *Bioscience Journal*, v.30, n.3, p.730-736, 2014.
- NAKASHIMA, Y.; SHIMIZU, K. Antitelmintic activity of *Thujopsis dolabrata* var. *Hondai*. III. Components with a termiticidal activity. *Miyazaki Daigaku Nogakubu Kenkyu Hokoku, Bulletin of the Faculty of Agriculture*, v.19, p.251-259, 1972.
- NTALLI, N.G.; CABONI, P. A review of isothiocyanates biofumigation activity on plant parasitic nematodes. *Phytochemistry Reviews*, v.16, n.5, p.827-834, 2017. <https://doi.org/10.1007/s11101-017-9491-7>
- QUIST, C.W.; SMANT, G.; HELDER, J. Evolution of plant parasitism in the phylum Nematoda. *Annual Review of Phytopathology*, v.53, n.1, p.289-310, 2015. <https://doi.org/10.1146/annurev-phyto-080614-120057>
- TARIQ, K.A.; CHISHTI, M.Z.; AHMAD, F.; SHAWL, A.S. Anthelmintic activity of extracts of *Artemisia absinthium* against ovine nematodes. *Veterinary Parasitology*, v.160, n.1-2, p.83-88, 2009. <https://doi.org/10.1016/j.vetpar.2008.10.084>
- VENTUROSO, L.R.; BACCHI, L.M.A.; GAVASSONI, W.L.; PONTIM, B.C.A.; CONUS, L.A. Influência de diferentes metodologias de esterilização sobre a atividade antifúngica de extratos aquosos de plantas medicinais. *Revista Brasileira de Plantas Medicinais*, v.12, n.4, p.499-505, 2010. <http://dx.doi.org/10.1590/S1516-05722010000400014>
- WIRATNO, W.; TANIWIRYONO, D.; VAN DEN BERG, H.; RIKSEN, J.A.G.; RIETJENS, I.M.C.M.; DJIWANTIA, S.R.; KAMMENGA, J.E.; MURK, A.J. Nematicidal activity of plant extracts against the root-knot nematode, *Meloidogyne incognita*. *The Open Natural Products Journal*, v.2, p.77-85, 2009. <https://doi.org/10.2174/1874848100902010077>

