Control of root rot (*Phytophthora cinnamomi*) in avocado (*Persea Americana*) with bioagents

Ciro Hideki Sumida[1], Lucas Henrique Fantin[1], Karla Braga[1], Marcelo Giovanetti Canteri[1], Martin Homechin (*in memorian*)

[1]Universidade Estadual de Londrina, Departamento de Agronomia. Rod. Celso Garcia Cid. (PR 445), Km 380, CP 6001, CEP 86051-990. Londrina, PR, Brasil.
Corresponding author: Karla Braga (karlabraga92@gmail.com)
Data de chegada: 27/02/2018. Aceito para publicação em: 19/12/2019.

10.1590/0100-5405/192195

ABSTRACT

Sumida, C.H.; Fantin, L.H.; Braga, K.; Canteri, M.G.; Homechin, M. Control of root rot (*Phytophthora cinnamomi*) in avocado (*Persea Americana*) with bioagents. ***Summa Phytopathologica,*** v.46, n.3, p.205-211, 2020.

Despite the favorable edaphoclimatic conditions for avocado production in Brazil, diseases such as root rot caused by the pathogen *Phytophthora cinnamomi* compromise the crop. With the aim of managing root rot in avocado, the present study aimed to evaluate chemical and biological control with isolates of *Trichoderma* spp. and *Pseudomonas fluorescens*. Thus, three assays were conducted to assess: (i) mycelial inhibition of *P. cinnamomi* by isolates of *Trichoderma* spp. and *P. fluorescens* from different crop systems; (ii) effect of autoclaved and non-autoclaved metabolites of *P. fluorescens*, and (iii) chemical or biological treatment of avocado seedlings on the control of root rot under field conditions. The isolates of *Trichoderma* spp. from maize cultivation soil and the commercial products formulated with *Trichoderma* presented greater antagonism ($p < 0.05$) to the pathogen *P. cinnamomi* in the *in vitro* tests. Similarly, non-autoclaved metabolites of *P. fluorescens* presented antagonistic potential to control *P. cinnamomi*. Under field conditions, the fungicide metalaxyl and the bioagents showed effectiveness in controlling *P. cinnamomi*, as well as greater root length and mass. Results demonstrated potential for the biological control of avocado root rot with *Trichoderma* spp. and *P. fluorescens.*

Keywords: Biological control, *Trichoderma* spp., *Pseudomonas fluorescens.*

RESUMO

Sumida, C.H.; Fantin, L.H.; Braga, K.; Canteri, M.G.; Homechin, M. Controle de podridão radicular (*Phytophthora cinnamomi*) do abacateiro (*Persea americana*) com bioagentes. ***Summa Phytopathologica,*** v.46, n.3, p.205-211, 2020.

Apesar das condições edafoclimáticas favoráveis para a produção de abacate no Brasil, doenças como a podridão da raiz causada pelo patógeno *Phytophthora cinnamomi*, comprometem a cultura. Visando o manejo da podridão radicular na cultura do abacateiro, o presente estudo teve como objetivo avaliar o controle químico e biológico com isolados de *Trichoderma* spp. e *Pseudomonas fluorescens*. Para isso foram conduzidos três ensaios, em que foram avaliados (i) a inibição micelial de *P. cinnamomi* por isolados de *Trichoderma* spp. e *P. fluorescens* provenientes de diferentes cultivos; (ii) efeito de metabólitos autoclavados e não autoclavados de *P. fluorescens* e (iii) o tratamento químico e biológico de mudas de abacateiro para o controle da podridão de raízes em condições de campo. Os isolados de *Trichoderma* spp. provenientes de solo de cultivo de milho e os produtos comerciais formulados com *Trichoderma* apresentaram antagonismo superior ($p<0,05$) ao patógeno *P. cinnamomi* nos testes "in vitro". Da mesma forma, metabólitos não autoclavados de *P. fluorescens* apresentaram potencial antagônico para o controle de *P. cinnamomi*. Em condições de campo, o fungicida metalaxil e os bioagentes apresentaram efetividade no controle de *P. cinnamomi* e maior comprimento e massa de raízes. Os resultados demonstraram o potencial do controle biológico da podridão radicular do abacateiro com *Trichoderma* spp. e *P. fluorescens*.

Palavras-chave: Controle biológico, *Trichoderma* spp, *Pseudomonas fluorescens.*

The avocado (*Persea americana* Mill.) is considered one of the main tropical fruits due to outstanding nutritional quality composed by omega fatty acids, phytosteroids, tocopherols and squalene; it is beneficial to health and can be used in consumption and for the pharmaceutical and cosmetics purposes (11). In the 2017 season, approximately 213,000 tons of fruit were produced in Brazil, with the State of São Paulo (49.8%), Minas Gerais (25.4%) and Paraná (10.4%) accounting for 85.7% of the country's production (15).

Despite the favorable edaphoclimatic conditions in Brazil, the production is limited due the many factors such as diseases. Root rot in the avocado is caused by the pathogen *Phytophthora cinnamomi* Rands. The disease is the main phytosanitary constraint in the avocado crop due to the susceptibility of main cultivars and the destructive power of pathogen (5, 23, 25). Among the management strategies are the use of tolerant rootstocks, acquisition or production of free pathogen seedlings, removal of cultural remains, planting in well-drained soils, balanced fertilization, as well as fungicide application and biological control (1).

Fungi of genus *Trichoderma* naturally inhabit the soil and can interfere the survival and activity of the plant pathogens, in addition to activating resistance mechanisms of the host plant (10, 4). In studies with *Trichoderma* sp. isolates, the results of Mcleod *et al*. (18) demonstrated effective control of *P. cinnamomi*. The same was presented in Cavero *et al*. (8) and Carvalho *et al*. (6) to the control of *Mycosphaerella fijiensis* Morelet and *Sclerotinia scleotiorum* (Lib.) de Bary, respectively. The potential of bacteria such as *Pseudomonas* sp. as a bioagent of plant pathogen control were observed. Studies showed the potential for control of lettuce root rot caused by *Pythium aphanidermatum* (Edson) Fitzp. (9) and can promote plant growth as well (28).

Thus, to evaluate the potential of biological agents as tools in the management of avocado root rot, the present study evaluated the effect of *Trichoderma* spp. and *Pseudomonas fluorescens* and their metabolites in the mycelial inhibition of *Phytophthora cinnamomi* and also the effect of the bioagents and fungicide in the control of the disease in the field.

MATERIAL AND METHODS

Three assays were developed. *In vitro* conditions, the first assay evaluated the mycelial inhibition of *P. cinnamomi* using *Trichoderma* spp. and *P. fluorescens* isolated from different culture systems (Table 1). The bioassay 2 evaluated the effect of autoclaved and non-autoclaved metabolites of *P. fluorescens* in antagonism to the pathogen *P. cinnamomi*. Bioassays 1 and 2 were carried out in the Phytopathology Laboratory of the State University of Londrina - PR. Assay 3 was conducted in a commercial area in the city of Cerqueira César - SP and evaluated the chemical and biological treatment with *Trichoderma* spp. and *P. fluorescens* of avocado seedlings in the control of root rot.

Isolation of *Phytophthora cinnamomi*

Phytophthora cinnamomi was isolated according to Zentmyer (32). Soil samples obtained from the rhizosphere of crop areas with plant with symptoms of root rot were added to a three-liter vessel, adding enough water to leave the aqueous layer at the bottom of the vessel. One avocado fruit, used as bait, was submerged in the soil for a period of four days at temperature approximately 28 ° C. The fruits were removed from the soil and washed in running water. Approximately 1 cm^2 of surface layer was removed and incubated in a moist chamber (gerbox 11x11x4cm) for 3 to 6 days. The colonies were identified based on the observation of reproductive and vegetative structures and then replace in V8-agar medium.

Assay 1- Biological control of *Phytophthora cinnamomi* with *Trichoderma* spp. and *Pseudomonas fluorescens* isolates

The assay was conducted "in vitro" by the culture technique cited by Mello *et al*. (19) in Petri dishes containing PDA (potato-dextrose-agar) medium. The design was completely randomized with five replicates and 16 treatments. The treatments were composed of ten isolates of *Trichoderma* spp. obtained from different crop systems, four *Trichoderma* spp. commercial products and two isolates of *P. fluorescens* from the collection of the Phytopathology Laboratory of Londrina State University, Londrina – Pr (Table 1).

Mycelial disks of the *P. cinnamomi* were arranged in plates with PDA culture media. The plates were kept in a climatic chamber at 23 ± 3ºC and photoperiod of 12 hours. The evaluation was based on the mycelial growth, which started four days after the initial of incubation and performed until the moment of control treatment reached the edge of the plates. The diameter of colonies of the pathogens was determined using a millimeter ruler. Mycelial growth inhibition was calculated using the formula:

% inib = [(crcontrol – crtreat) /crcontrol] x 100

where, **crcontrol** is the mycelial growth of control and **crtreat** corresponds to the mycelial growth of treatment.

Table 1. Source of isolates of *Trichoderma* spp., *Pseudomonas fluorescens* and commercial products based on genera *Trichoderma*.

Isolate	Crop system	Source
Maize DP5*	Maize	Londrina-PR
Maize DP6*	Maize	Londrina-PR
Maize DA5*	Maize	Londrina-PR
Maize DA2*	Maize	Londrina-PR
Maize CP4*	Maize	Londrina-PR
Avocado	Avocado	Piraju-SP
Coconut	Coconut	Mauá da Serra-PR
Persimmon	Persimmon	Mauá da Serra-PR
Sugarcane	Sugarcane	Londrina-PR
Coffee	Coffee	Londrina-PR
C1	Londrina State University	Londrina-PR
C2	Londrina State University	Londrina-PR
Agrotrich®	Commercial product	-
Trich Organic®	Commercial product	-
Trichodermil®	Commercial product	-
Biotrich®	Commercial product	-

*DA5 - no-tillage system after oats; DA2 - no-tillage system after oats ; DP5 - no-tillage system after fodder; DP6 - no-tillage system after fodder and CP4 - conventional planting after fallow

Assay 2- Metabolites of *Pseudomonas fluorescens* in antagonism to *Phytophthora cinnamomi*

The effect of autoclaved and non-autoclaved metabolites of *P. fluorescens* in antagonism to *P. cinnamomi* was assessed. The design was completely randomized with seven treatments and five replications. The treatments consisted of the metabolites produced by the isolates C1, C2 and C1 + C2, submitted or not to the autoclaving process versus the test with only *P. cinnamomi*. The metabolites were obtained from *P. fluorescens* from the Laboratory of Phytopathology of Londrina State University, Londrina - Pr, multiplied in Potato dextrose medium under agitation for 36 hours, followed by vacuum filtration with filter paper. Subsequently, the medium was poured into the plates, and a disc was deposited at the center of each plate (5 mm diameter) from *P. cinnamomi* colonies from the bait isolation. The mycelial inhibition followed the procedures described in assay 1.

Assay 3- Chemical and Biological treatment of avocado seedlings inoculated with *Phytophthora cinnamomi*

The assay was conducted in a commercial area in the city of Cerqueira César - SP. The seedlings of cultivar "Margarida" were obtained by sowing in plastic bags containing a substrate composed of a mixture of 3.0 kg of single superphosphate, 0.5 kg of potassium chloride and 100 liters of cattle manure for each cubic meter of soil of ravine. Previously, the substrate was sterilized to ensure not contamination. The solution of inoculum of *P. cinnamomi* was obtained by the crushing of 500g of roots of symptomatic plants (Figure 1) per liter of water. The solution was applied in the colon region of seedlings

Figure 1. Symptomatic avocado trees with Root Rot - Fazenda Jurumirim- Cerqueira César-SP.

with 30 days (50mL.plant^{-1}). The experiment was accomplished at a nursery (50% light). The experimental design was completely randomized with four replicates (seedlings) and 10 treatments.

The biological treatments were composed by the solutions with *Trichoderma* spp. (persimmon, MaizeCP4, MaizeDP5 and Trichodermil®), and *P. fluorescens* (C1 and C2). For the chemical treatment were used fungicide Ridomil® (metalaxil) and the fertilizer Hortifós®. The biological treatment consisted of applications in the stem close to root. For this, 5.0 mL *Trichoderma* spp. suspension, at the concentration of 1.10^4 spores. mL^{-1} and 5.0 mL of *P. fluorescens* suspension were at approximately 120 cfu.mL^{-1} (colony forming unit) were applied. For the chemical treatments, the solution applied was 0.5 mL of the Hortifós® fertilizer, diluted in 4.5 mL of water, and sprayed onto the leaves. The fungicide Ridomil® was applied to the soil close to stem plant, in the proportion of 0.5 g of commercial product, diluted in 5 ml of water per seedling. The evaluations were performed at 120 days after application. The evaluated parameters were leaf and root state, plant height and root length, fresh and dry weight of shoot and roots. Leaf and root state were rated according to the characteristics presented by the free control of the pathogen without inoculation (NI) and without treatment (NT).

Statistical analysis

The data were submitted to analysis of variance (ANOVA) and F test, with a significance level of 0.05. The treatments were compared by the Tukey test. Statistical analyzes were performed using the R program (26).

RESULTS AND DISCUSSION

Biological control of *Phytophthora cinnamomi* with *Trichoderma* spp. and *Pseudomonas fluorescens* isolates

The mycelial inhibition of *P. cinnamomi* due to treatment with *Trichoderma* spp. (Maize DP5 = 73.3% and Maize CP4 = 73.3%) and commercial products Trichodermil®, Agrotrich® and Trich Organic® presented inhibition control higher than 70% and higher than *P. fluorescens* isolates (C1 and C2) as well. The lowest level of control was observed for isolates from soils cultivated with sugarcane, with 24.0% inhibition (Figure 2).

The inhibition promote by the treatments with *P. fluorescens* were 40.0 and 44.0%. The results were superior to those presented by the treatments with *Trichoderma* spp. isolated from coffee, sugarcane

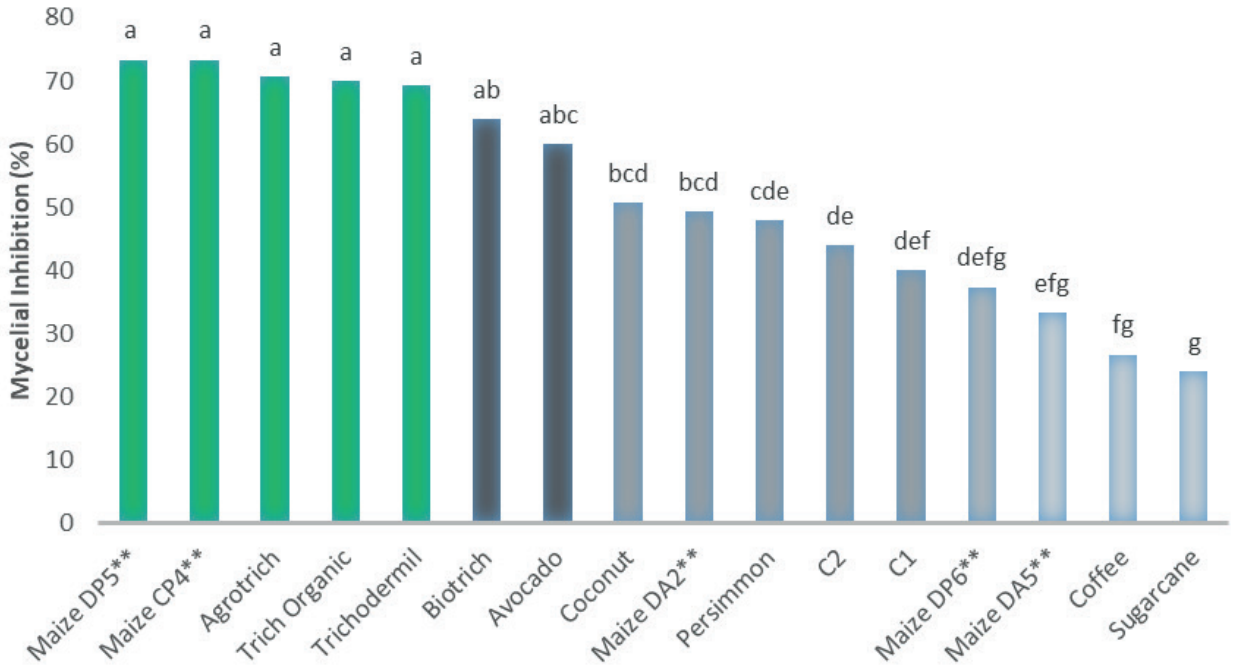

Figure 2. Mycelial Inhibition of *Phytophthora cinnamomi* by *Trichoderma* spp. and *Pseudomonas fluorescens* isolates. ** DA = direct seeding after oats; DP = no-till after no-till; CP = conventional sowing after fallow. Means followed by the same letter do not differ by the Tukey test at the 5% level of significance.

and no-tillage systems of maize after fallow and oats (maizeDP6 and maizeDA5). In vitro, Méndez-Bravo *et al* (21) studying the antagonist potential of rhizobacterial isolates from asymptomatic and symptomatic avocado root rot showed inhibition of *P. cinnamomi* micelial growth of 46% by *Bacillus acidiceler*. The *Pseudomonas baetica* isolate presented inhibition less than 10%.

Metabolites of *Pseudomonas fluorescens* in antagonism to *Phytophthora cinnamomi*

The evaluations were performed when the mycelial growth of the control reached the edge of the plaque, approximately 96 hours after inoculation (Figure 3).

The results showed mycelial inhibition for the treatment with autoclaved metabolites T1 (isolate C1), T2 (isolate C2) and T3 (mixture C1 + C2) less than non-autoclaved T4 (isolate C1), T5 (isolate C2) and T6 (mixture C1 + C2) (Figure 4). The inhibition of non-autoclaved metabolites was higher than 90%. Autoclaved metabolites produced by isolate C1 presented less inhibition control (39 %). The mixture of isolates C1 and C2 did not presented increases in inhibition control of *P. cinnamomi*. Working with non-autoclaved metabolites of bacterial isolates obtained from avocado rhizosphere, Ramírez *et al*. (27) showed inhibitory activity against the evaluated pathogens *P. cinnamomi, Colletotrichum* sp. and *Phomopsis* sp.

The reduction of effectiveness of autoclaved compounds can be explained by the volatility of compounds produced by the bacteria. Méndez-Bravo *et al* (21) characterized metabolites produced by bacterial isolates of genera *Bacillus, Pseudomonas* and *Arthrobacter* from avocado trees rhizosphere. The authors showed that metabolites are composed by diffusible and volatile parts, and the main volatile compounds produced by the *Bacillus* were ketones, aldehydes, alkyls, sulfoxides, pyrazines and alcohols. In addition, the bacteria metabolites varying according to genera and species.

Bacterial metabolites have a great potential to control phytopathogens. The characterization of metabolites was studied in many pathosystems, such as the *P. infestans* growth and sporulation was inhibited by compounds Dimethyl disulfide and other sulfur-containing compounds produced by *Pseudomonas* (31) as well as the effectiveness of other volatile compounds was presented in the control of *Botrytis cinerea, Penicillium digitatum* and *P. italicum* (14).

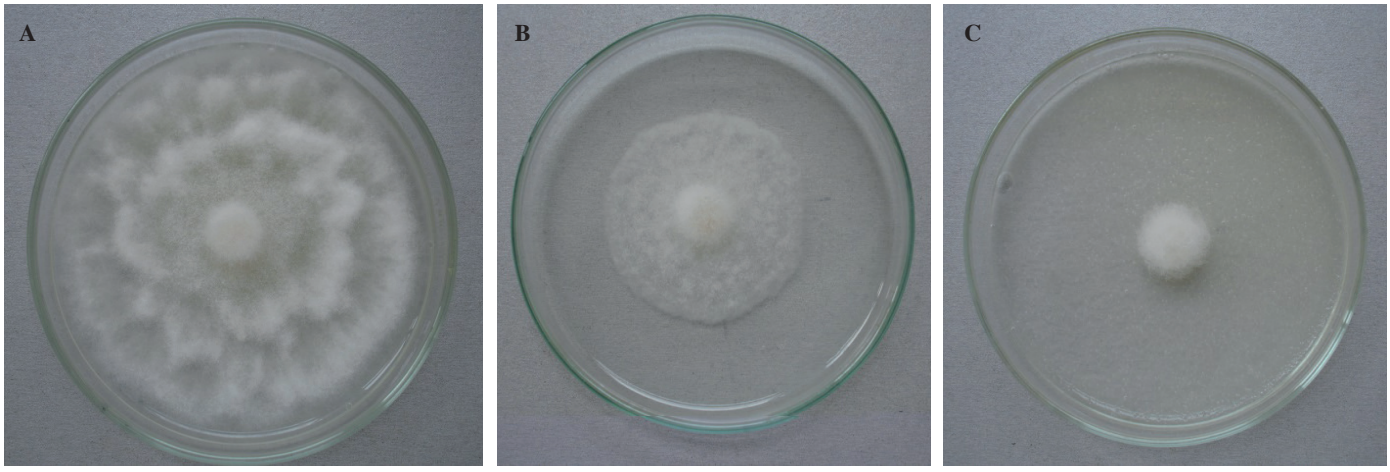

Figure 3. Mycelial Inhibition of *Phytophthora cinnamomi* submitted to autoclaved and non-autoclaved *Pseudomonas fluorescens* metabolites 96 hours after inoculation. (**A**) control treatment without metabolites; (**B**) treatment "T1" autoclaved metabolite and (**C**) treatment "T4" non-autoclaved metabolite.

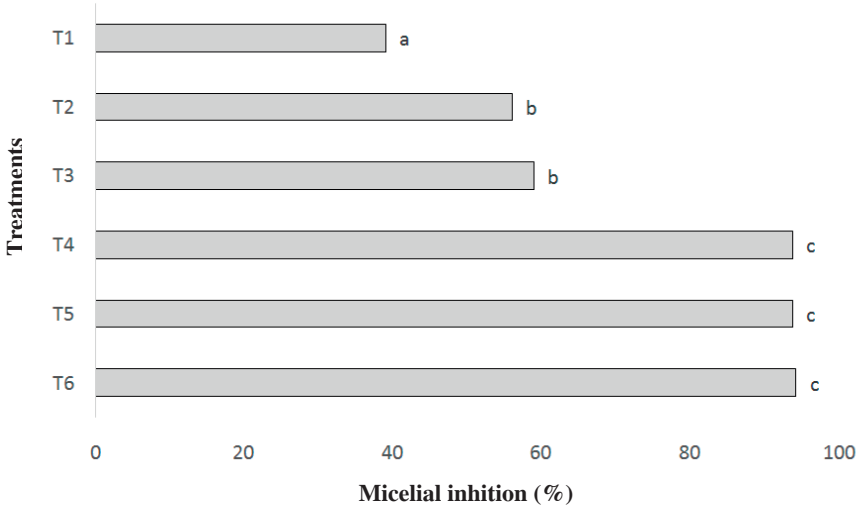

Figure 4. Inhibition of growth of *Phytophthora cinnamomi* in BDA medium containing *Pseudomonas fluorescens* Means followed by the same letter do not differ by the Tukey test at the 5% level of significance. T1, T2 and T3 were autoclaved, T4, T5 and T6 were not autoclaved.

Figure 5. Evaluation of roots of seedlings performed at 120 days after inoculation of *Phytophthora cinnamomi.* (**A**) control without inoculation; (**B**) treatment with application of fungicide Ridomil® (**B**) and with *Trichoderma* spp. isolated from soil with persimmon (**C**).

The results of present study indicated potential of use of metabolites in the control of rot roots in avocado and more studies are necessaries to characterize metabolites produced and the response of them to factors such as temperature, light, etc.

Chemical and Biological treatment of avocado seedlings inoculated with *Phytophthora cinnamomi*

There was a reduction in seedling development in treatment with inoculation of *P. cinnamomi* and without application (NT) (Figure 5).

The treatment with fungicide Ridomil® showed higher development of seedlings for the parameters height, fresh weight, plant dry weight and normal staining; but with regular root length, not differing from other treatments (Table 2).

The results obtained, corroborate those of Leal *et al*. (16). which observed a reduction in disease severity and in the *P. cinnamomi* population in the soil, as well as an increase in height and dry weight of the seedlings treated with Ridomil®. Gonzalez e at (12) showed reduced of growth colony of *P. cinnamomi* of oak higher than 75% with metalaxyl application. The authors also presented reduced of root lesions in plants with metalaxyl or Potassium phosphite application.

On the other hand, the seedlings treated with *Trichoderma* from "persimmon" soil (Figure 5C) presented the highest root length, but absence of radicles may compromise the future development of the seedlings due to less capacity of water and nutrient absorption. Different results were present for Stefanello *et al*. (29), that using *Trichoderma harzianum* in the control of root rot of cassava, observed increased productivity for the treatments with the use of bioagent. Sumida *et al*. (30) and Haddad *et al*. (13) also observed an antagonistic reaction of *Trichoderma* to isolates of *Sclerotinia sclerotiorum*, that causes white mold. Méndez-Bravo *et al* (21) demonstrated that volatile compounds

Table 2. Evaluation of the development of avocado seedlings, inoculated with *Phytophthora cinnamomi* and treated with bioagents *Trichoderma* spp. and *Pseudomonas fluorescens* in commercial area.

Tratamentos	Root				Aerial			
	lenght (cm)	WDM (g)	FW (g)	State	lenght (cm)	WDM (g)	FW (g)	State
C. NI and NT	57,5 ab	57,25 b	138,5 d	normal	56,25 a	42 abc	83,5 abc	normal
C. WI e NT	45 a	22,25 a	46,5 a	Sympt.	41 a	19,5 a	35,25 a	Sympt.
Milho DP5	57,75 ab	24,5 a	66,5 ab	normal	48,75 a	27,5 ab	52 ab	Sympt.
Tricodermil	49,5 a	49,5 ab	123,5 cd	normal	59,5 a	50,5 abc	86 abc	normal
Maize CP4	47,75 a	28,5 a	60,5 ab	Sympt.	47,75 ab	28,5 bc	60,5 bc	Sympt.
Persimmon	71,33 b	29,33 ab	51,5 a	Sympt.	44 a	29 ab	64,7 ab	normal
C1	47,5 a	33,5 ab	77,5 abc	normal	61 ab	51,5 abc	86,5 abc	normal
C2	56 ab	31,5 ab	91,5 abcd	Sympt.	57,75 a	37 ab	64 ab	Sympt.
Hortifós	53,5 ab	26,5 a	55,5 a	normal	65,25 ab	36,5 ab	68 ab	normal
Ridomil	54,5 ab	35,4 ab	115,5 cd	normal	87,5 b	76 c	128 c	normal

Means followed by the same letter in the columns do not differ from each other to a 5% significance level by the Tukey test; C. NI and NT = Control without inoculation and treatment; C. WI and NT = Control with Inoculation and no Treatment; lenght (cm) = length in centimeters; WDM (g) = weight of dry matter in grams; FW (g) = fresh weight in grams, State = situation of root; Sympt = presence of symptoms., normal= No presence of symptoms.

produced by bacterial isolates from rhizosphere showed biocontrol of soil borne oomycetes such as *P. cinnamomi* and the antagonism can be related to metabolites produced by bioagents.

Despite of present study showed higher efficiency control of *P. cinnamomi* in seedlings of the chemical control than biological agents, the results demonstrated potential of biological control of avocado root rot. The benefits of bioagents uses are beyond the pathogen inhibition. The use of biological agents or plant extracts may contribute to delay the development of fungicide resistance (7). Marcías-Rodríguez *et al*. (17) explain that *Trichoderma atroviride* relationship with tomato plant promoted tomato development and changes in root carbohydrates exudation, consequently provides favorable conditions to the Trichoderma growth and antagonism against *P. cinnamomi*.

In general, the bioagents that survives in plant rhizosphere demonstrated potential to be used in root rot management systems caused by soil pathogens. The results alert the importance of measures to ensure the maintenance of bacterial and fungal community rhizosphere in the equilibrium of crop systems. In this context, more studies of combinations of integrated management such as biofumigation with pellets of plants (22), use of natural essential oils (24), natural plant extracts (2), resistance cultivars (3, 31), chemical fungicides and fertilizers such as Potassium phosphite (12) are necessary.

The *Trichoderma* spp. isolates from maize soil system and commercial biological products (Trichodermil®, Agrotrich® and Trich Organic ®) presented antagonism against the pathogen *P. cinnamomi* in the in vitro tests. In the same way, non-autoclaved metabolites of *P. fluorescens* demonstrate potential antagonistic against *P. cinnamomi*. In field conditions, the fungicide Ridomil® and the bioagents showed effectiveness in controlling *P. cinnamomi* and greater length and root mass. The results demonstrated potential of biological control of avocado root rot with *Trichoderma* spp. and *P. fluorescens*.

ACKNOWLEDGEMENTS

The authors are grateful for the financial support of the CNPq and Araucária Foundation for Scientific and Technological Development (Paraná, Brazil).

REFERENCES

1. Amorim, L.; Rezende, J. A. M.; Bergamin Filho, A.; Camargo, L. E. A. **Manual de fitopatologia:** doenças das plantas cultivadas. 5. ed. Ouro Fino: Agronômica Ceres, 2016. v. 2, 772 p.
2. Arroyo Cordero, F. T.; Rodríguez-Arcos, R.; Jiménez-Araujo, A.; Guillén--Bejarano, R.; Basallote, M. J.; Barrau, C. Inhibitory effect of the glucosinolate–myrosinase system on *Phytophthora cinnamomi* and *Pythium spiculum*. **Plant Protection Science**, Praga, v. 55, p. 93-101. 2019.
3. Belisle, R. J.; McKee, B.; Hao, W.; Crowley, M.; Arpaia, M. L.; Miles, T. D.; Adaskaveg, J. E.; Manosalva, P. Phenotypic Characterization of Genetically Distinct *Phytophthora cinnamomi* Isolates from Avocado. **Phytopathology**, St. Paul, v.109, p. 384-394. 2019.
4. Benitez, T.; Rincon, A. M.; Limón, M. C.; Códon, A. C. Biocontrol mechanisms of *Trichoderma* Strains. **International Microbiology**, Madrid, v. 7, p. 249-260, 2004.
5. Cantuarias-Avilés, T.; Silva, S. R. The avocado industry in the state of Sao Paulo, Brazil: present and future perspectives. In: Congreso Mundial Del Aguacate, 7., 2011, Cairns: Avocado Source, 2011.
6. Carvalho, D. D. C.; Geraldine, A. M.; Lobo Junior, M.; de Mello, S. C. M. Biological control of white mold by *Trichoderma harzianum* in common bean under field conditions. **Pesquisa Agropecuária Brasileira**, Brasília, v. 50, n. 12, p. 1220-1224, 2015.
7. Castillo-Reyes, F.; Hernández-Castillo, F. D.; Clemente-Constantino, J. A.; Gallegos-morales, G.; Rodríguez-Herrera, R.; Aguilar, C. N. In vitro antifungal activity of polyphenols-rich plant extracts against *Phytophthora cinnamomi* Rands. **African Journal of Agricultural Research**, Nigéria, v. 10 n. 50, p. 4554-4560, 2015.
8. Cavero, P. A. S.; Hanada, R. E.; Gasparotto, L.; Coelho Neto, R. A.; Souza, J. T. Controle biológico da Sigatoka-negra da bananeira com *Trichoderma*. **Ciência Rural**, Santa Maria, vol.45, n.6, p.951-957, 2015.
9. Cipriano, M. A. P.; Patrício, F. R. A.; Freitas, S. S. Potencial de rizobactérias na promoção de crescimento e controle da podridão radicular em alface hidropônica. **Summa Phytopathologica**, Botucatu, v.39, n.1, p.51-57, 2013.
10. Cook, R. J.; Baker, K. F. **The nature and practice of biological control of plant pathogens**. St. Paul: APS, 1983. 539 p.
11. Duarte, P. F.; Chaves, M. A.; Borges, C. D.; Mendonça, C. R. B. Avocado: characteristics, health benefits and uses. **Ciência Rural**, Santa Maria, v.46, n.4, p.747-754, 2016.
12. González , M.; Caetano, P.; Sánchez, M. E. Testing systemic fungicides for control of *Phytophthora* oak root disease. **Forest Pathology**, Hoboken, v. 47, n. 4, 2017.
13. Haddad, P. E.; Leite, L. G.; Lucon, C. M. M.; Harakawa, R. Selection of *Trichoderma spp.* strains for the control of *Sclerotinia sclerotiorum* in soybean. **Pesquisa Agropecuária Brasileira**, Brasília, v. 52, n. 12, p. 1140-1148, 2017.
14. Huang, R.; Li, G. Q.; Zhang, J.; Yang, L.; Che, H. J.; Jiang, D. H.; Huang, H. C. Control of postharvest Botrytis fruit rot of strawberry by volatile organic compounds of *Candida intermedia*. **Phytopathology**. St. Paul, v.101, n.7, p. 859-69. 2011.
15. Instituto Brasileiro de Geografia e Estatística. Produção Agrícola - Lavoura Permanente. 2018. Disponível em: https://cidades.ibge.gov.br/brasil/pesquisa/15/0. Acesso em: 07 fev. 2018.
16. Leal, J. M.; Castaño, J.; Bolaños, M. M. Manejo de la pudrición radical (*Phytophthora cinnamomi* RANDS) del aguacate (*Persea americana* LINNEO). **Revista U.D.C.A Actualidad & Divulgación Científica**, Bogotá, v.17, n.1, p.105-114, 2014.
17. Macías-Rodríguez, L.; Guzmán-Gómez, A.; García-Juárez, P.; Contreras--Cornejo, H. A. *Trichoderma atroviride* promotes tomato development and alters the root exudation of carbohydrates, which stimulates fungal growth and the biocontrol of the phytopathogen *Phytophthora cinnamomi* in a tripartite interaction system. **FEMS Microbiology Ecology**, Oxford, v.94, n.9, 2018.
18. Mcleod, A.; Labuschagne, N.; Kotzé; Johannes, M. Evaluation of *Trichoderma* for biological control of avocado root rot in bark medium artificially infested with *Phytophthora cinnamomi*. **South African Avocado Growers' Association Yearbook**, v. 18, p. 32-37, 1995.
19. Mello, S. C. M.; Ávila, Z. R.; Braúna, L. M.; Pádua, R. R.; Gomes, D. Cepas de *Trichoderma* para el control biológico de *Sclerotium rolfsii* Sacc. **Fitosanidad**, La Habana, v. 11, n. 1, p. 3-9, 2007.
20. Melo, I. S. Potencialidades de utilização de *Trichoderma spp*. no controle biológico de doenças de plantas. In: Bettiol, W. **Controle biológico de doenças de plantas**. Campinas: EMBRAPA/CNPDA, 1991. p. 135-156.
21. Méndez-Bravo, A.; Cortazar-Murillo, E. M.; Guevara-Avendaño, E.; Ceballos-Luna, O.; Rodríguez-Haas, B.; Kiel-Martínez, A. L.; Hernández-Cristóbal, O.; Guerrero-Analco, J. A.; Reverchon, F. Plant growth-promoting rhizobacteria associated with avocado display antagonistic activity against *Phytophthora cinnamomi* through volatile emissions. **Plos One**, San Francisco, v.13, n.3, p. e0194665. 2018.
22. Morales-Rodríguez,C.; Vettraino, A. M.; Vannini,A. Efficacy of Biofumigation with *Brassica carinata* Commercial Pellets (BioFence) to Control Vegetative and Reproductive Structures of *Phytophthora cinnamomic*. **Plant Disease**, St. Paul, v.100, n.2, p.324-330. 2016.
23. Nakasone. H. Y.; Paull. R. E. **Tropical Fruits**. In: **Crop production science in horticulture.** 2. ed. Wallingford: CABI, 1998. p. 95.
24. Ochoa Fuentes, Y.; Hernández Pérez, A.; Delgado Ortiz, J.; Cerna Chavez, E.; Aguirre Uribe, L.; Tapia-Vargas, L. Control orgánico in vitro de *Phytophthora cinnamomi* con aceites esenciales de orégano y clavo. **Revista Mexicana de Ciencias Agrícolas**, Texcoco, v. 10, n. 4, p. 961-968. 2019.
25. Pérez - Jiménez R. M. Significant Avocado Diseases Caused by Fungi and Oomycetes. **The European Journal of Plant Science and Biotechnology**, Tokyo, v. 21, p. 1-24, 2008.
26. R Core Team. A language and environment for statistical computing. Vienna: R Foundation for Statistical Computing, 2017. Available: https://www.R-project.org/.

27. Ramírez Restrepo, S; Arias Marin, J. D.; Bedoya Perez, J. C.; Rueda Lorza, E. A.; Yaneth Sánchez, C.; Granada García, S. D. Metabolites produced by antagonistic microbes inhibit the principal avocado pathogens in vitro. **Agronomía Colombiana**, Bogotá, v. 33 n. 1, p. 58-63, 2015.
28. Robbs, C. E. Bactérias como agentes de controle biológico de fitopatógenos. In: Bettiol, W. **Controle biológico de doenças de plantas**. Campinas: EMBRAPA/CNPDA, 1991. p. l21-133.
29. Stefanello, L.; Stefanello, V. F. V.; Heling, A. L.; Henkemeier, N. P.; Roncato, S. C.; Kuhn, O. J.; Stangarlin, J. R. Manejo da podridão radicular da mandioca pela combinação de manejo de solo, variedade resistente e controle biológico com *Trichoderma harzianum*. **Raízes e Amidos Tropicais**, Botucatu, v. 13, n. 1, p. 31-45, 2017.
30. Sumida, C. H.; Daniel, J. F. S.; Araújo de, A. P. C. S.; Peitl, D. C.; Abreu, L. M.; Dekker, R. F. H.; Canteri, M. G. *Trichoderma asperelloides* antagonism to nine *Sclerotinia sclerotiorum* strains and biological control of white mold disease in soybean plants. **Biocontrol Science and Technology**, London, v. 28, n. 2, p. 142-156, 2018.
31. De Vrieze, M.; Pandey, P.; Bucheli, T. D.; Varadarajan, A. R.; Ahrens, C. H.; Weisskopf, L.; Bailly, A. Volatile Organic Compounds from Native Potato-associated Pseudomonas as Potential Anti-oomycete Agents. **Frontiers in Microbiology**, Rockville, v.6, p.1295. 2015.
32. Zentmyer, G. A. ***Phytophthora cinnamomi*** **and the diseases it causes**. Monograph n° 10. St. Paul: The American Phytopathological Society, 1980. 96p.