Journal of Population Therapeutics & Clinical Pharmacology

RESEARCH ARTICLE
DOI: 10.53555/jptcp.v30i8.3120

IN VITRO ANTIFUNGAL ACTIVITY OF SWINGLEA GLUTINOSA LEAF EXTRACT AGAINST *COLLETOTRICHUM GLOEOSPORIOIDES* ON *PERSEA AMERICANA* MILL

Pavel Peroza-Piñere[1*], Donicer E. Montes Vergara[1], Alexander Pérez Cordero[1]

[1*]Universidad de Sucre, Facultad de Ciencias Agropecuarias, Colombia, pavel@fca.edu, https://orcid.org/0000-0002-6726-3871
[1]donicer.montes@unisucre.edu.co, https://orcid.org/0000-0002-2860-0505
[1]alexander.perez@unisucre.edu.co, https://orcid.org/0000-0003-3989-1747

***Corresponding author :** alexander.perez@unisucre.edu.co
*Universidad de Sucre, Facultad de Ciencias Agropecuarias, Sincelejo, Sucre, Colombia

Submitted: 22 September 2022 **Accepted:** 5 December 2022 **Published:** 30April 2023

ABSTRACT

The avocado (*Persea americana* Mill.) is a species whose cultivation is of great nutritional and economic importance for the Maria mounts region, the variety of avocado grown is the Antillean race and var. lorena; however, like any other crop, it is often affected by pests and diseases that limit its commercialization at national and world level. The phytopathogenic fungus *Colletotrichum gloeosporioides* is the causal agent of anthracnose in avocado and manifests itself in the early stages of fruit development, as well as in postharvest and during storage, under conditions of high relative humidity (80%) and temperatures from 20 ◦C. Economic losses due to this fungus can be up to 20% of production. Therefore, the objective of this work was to evaluate the antifungal activity of extracts and fractions of *Swinglea glutinosa* against *Colletotrichum gloeosporioides*, in avocado crop The plant material used corresponded to dried leaves of *S. glutinosa* collected in two agroecological regions of the department of Sucre, Colombia. The collected plant material was used to obtain ethanolic extracts. Three chromatographic fractions were obtained from the extracts and four concentrations (1%, 25%, 50% and 75%) were prepared per municipality. Each concentration was used for *In vitro* evaluation against *C. gloeosporioides*. The results obtained show that at a concentration of 75%, the highest percentages of antifungal index (% A.I.) were obtained for the bioproduct fraction 2 and extract from the municipality of Sincelejo with a % A.I. of 96.6% and 87.2%, respectively. In this study, both extracts and fractions 2 showed 100% inhibition in the germination of *C. gloeosporioides* conidia at a concentration of 25%. The chromatographic profile of the bioproducts found evidenced the presence of terpenoid compounds, in which antimicrobial activity has been demonstrated.

Keywords: phytopathogens, bioproducts, anthracnose, avocado, *Colletotrichum gloeosporioides*

INTRODUCTION

The Avocado also known as avocado (Hurtado, Fernandez & Carrasco, 2018), has been experiencing an increase in demand in international markets, which contrasts with the growing but deficient supply by producing countries that do not meet the consumption needs of importing countries Ramirez, Gonzalo & Peterson, (2018).

Anthracnose is a disease generated by the fungus *Colletotrichum gloeosporioides*, the microorganism in nature that lives on matter, attacks very young tissues or very old and physically weak tissues of plants. The most severe attacks on fruit occur when the most susceptible crop stage (flowering or fruiting) coincides with rainy weather and days of permanent relative humidity above 90%. Inoculum sources are found on leaves, branches, inflorescences, flower bracts and fruit, in general terms throughout the tree (Alarcón and Chavarriaga, 2007).

Among the various alternatives, the use of plant-based products, which are biodegradable, has attracted the attention of scientists around the world. Plant-derived products, besides being economical and environmentally safe, have proven to be effective in controlling post-harvest diseases and are therefore considered ideal candidates for use as agrochemicals (Macias et al., 1997). Lemon and papaya leaf extracts have been used for the control of other post-harvest fungi, such as *Fusarium* spp*., Alternaria* spp*., Pestalotiopsis* spp*. and Rhizopus* spp*. C. papaya* leaf extract completely inhibited *In vitro* rots caused by *C. gloeosporioides*. In Colombia, the fungicidal effect of *C. papaya* on *Mycosphaerella fijiensis* Morelet was evaluated using castor oil as an adherent, and the authors found that there were sensitivity responses to the fungus (Marín et al., 2008).

S. glutinosa (Blanca) Merr. (Rutaceae), popularly known as Ornamental Lemon or Tabog, is a native species of the Philippines. It was introduced to Panama in the 1930s at the Summit Botanical Garden as part of a collection of rare citrus species from tropical Asia. Its introduction was made in order to have genetic materials for citrus breeding and also for use as a resistant rootstock for grafting fruiting citrus (Stashenko et al., 2015).

Swinglea extract is a botanical fungicide obtained from plant extracts, rapidly biodegradable and harmless to man and the environment, developed to prevent and cure diseases in flowers, fruit trees, vegetables, ornamental plants and beans. It is composed of 80% *swinglea* plant extracts, which also include Eugenol, Linalyl, Citrolenol, Geraniol and terpinen. It has a pH of 4.5, and acts as a fungicide and protectant. The extract has been used for the control of anthracnose in some species and for powdery mildew and wilt in beans (Abonos Superior Ltda. 2005). Torres-Gonzalez (2013) reported that this extract contains alkaloids, terpenoids and flavonoids, metabolites with fungicidal effect. The commercial product is classified as low toxicity.

Several products of plant origin have demonstrated an antimicrobial effect. These compounds include flavonoids, phenols, terpenes, essential oils, alkaloids, lectins and polypeptides (Cowan, 1999). Rao et al. (1992) separated and evaluated the fungistatic effect of essential oils from various plants, and found the greatest effectiveness against *C. gloeosporioides*, *Curvularia pallescens* and *Periconia atrovirens*, with those extracted from the flowers of *C. cyminum* Mill and *S. aromaticum* L. Both plants showed a fungistatic effect at 1000 ppm and a fungicidal effect at 2000 and 3000ppm. They also determined that the fungitoxic effect of mineral oil resided in its aldehyde and phenolic fractions. In this sense, products of plant origin such as extracts and essential oils of citrus and aromatic plants have been considered for the control of phytopathogenic fungi both at harvest and postharvest (Landero-Valenzuela, 2016). The latter stage is critical, so essential oils with antifungal effect constitute an alternative as an inhibitory component in edible coating formulations designed to reduce spoilage and fungal infections at the postharvest stage (Rimá de Oliverira et al., 2017). *Swinglea glutinosa* Merr is a plant species, commonly known as fence lemon, belonging to the Rutaceae family and recognized for its use in the formation of living fences (Cifuentes et al., 2019); in addition, the extract of this ornamental has been reported to control weeds (Gil et al., 2010) and some fungal diseases in certain crops (Hincapié et al., 2017).

S. glutinosa is immune to *Phytophthora* foot rot or gummosis. Field observations indicate that it is also highly resistant to citrus tristeza, caused by a virus. Other species of the Rutaceae family do not exhibit such resistance (Rondón, 2008). The action of some secondary metabolites present in this shrub could explain the evident resistance of this shrub to the action of these pathogens against other species of the same family; the metabolites responsible for this effect are the following

Taking into account the previous problem, the research and use of natural resources is justified, to be used as biological potential for the benefit of society, conducting studies that allow the discovery of new bioactive molecules, which can be found from plant sources, such as lemon *S. glutinosa*, whose biological potential is unknown in terms of inhibition of disease-causing microorganisms in the avocado crop in the sub-region of Maria Mounts, north coast of Colombia. The present research aims to evaluate the *in vitro* antifungal effect of *S. glutinosa* plant extract on the phytopathogenic fungus *Colletotrichum* sp.

MATERIAL AND METHODS

Study site and sampling. Samples of *Swinglea glutinosa* were collected in the department of Sucre, Colombia in the municipality of Sincelejo (Montes de María subregion) at 9° 17' 12.82" N and 75° 23' 55.83" W at 187 masl and Santiago de Tolú (Gulf of Morrosquillo subregion) at 9° 29' 57.57" N and 75° 35' 36.40" W at 3 masl. The fungus used for the antifungal assays was isolated from avocado (*Persea americana*) leaves and fruits in the Montes de María subregion. The fruits were collected during the fruiting season, at the physiological maturity stage, from farms in the municipality of Chalán (9°32'38" N and 75°18'45" W, at an altitude of 408 m.

Obtaining fractions by column chromatography. The previously rotated ethanolic extracts were taken to a glass chromatographic column with bulb previously packed with silica gel 60 (0.063-0.200 mm) as stationary phase and as a gradient mobile phase the solvent mixtures (F1) Cyclohexane: Trichloroethylene (6:4), (F2) Chloroform: Acetone (8:2) and (F3) Methanol, obtaining three fractions per extract.

Mycelial growth inhibition. For the mycelial growth inhibition test, the direct seeding methodology proposed by Pérez et al. (2021) was followed with modifications. In this test, isolates of approximately 7 mm diameter growth area of *Colletotrichum gloeosporioides* were used and seeded on the surface of PDA medium enriched with chloramphenicol, rifampicin and ampicillin. The two total extracts and six fractions were evaluated for a total of 8 bioproducts, preparing concentrations at 1%, 25%, 50% and 75% in Tween 20 to 1% solution and 0.2% DMSO (Espitia et al., 2014), Benomil 2% negative control, negative control Tween 20 1% solution and 0.2% DMSO and an absolute control was used as positive control. From each treatment, 5 replicates were performed for a total of 175 experimental units.

Minimum inhibitory concentration (MIC) and minimum fungicide concentration (MFC). With the most efficient bioproducts, concentrations of 75%, 80%, 85%, 90%, 95% and 100% dissolved in 1% Tween 20 and 0.2% DMSO were prepared; a negative control and an absolute control were used. Five replicates per treatment were performed for a total of 130 experimental units.
MIC and MFC values were calculated on mycelial growth on PDA enriched with chloramphenicol, rifampicin and ampicillin and incubated at 28±2°C taking into account the absolute control and the negative control. The MIC of extracts and fractions was defined as the lowest concentration of treatments that showed growth inhibition ≥90% at 8 days of incubation (Consentino et al., 1994). The MFC of extracts and fractions was defined as the lowest concentration of treatments that showed no visible fungal growth or caused growth inhibition around 100% at 14 days of incubation.

Germination curve of *Colletotrichum gloeosporioides conidia*. This test was performed to determine the maximum germination period of the *C. gloeosporioides* strain using a 1% solution of Tween 20 and 0.85% NaCl in sterile distilled water as substrate. To develop this test, the fungus strain was incubated for 15 days in PDA medium with intervals of 12 hours of light and 12 hours of darkness.

Statistical analysis. The research was carried out under a DBCA 4x4 design with 5 replicates per treatment and an ANOVA statistical analysis with simple factorial arrangement with sampling in the experimental units, using the statistical program Statgraphics Centurión XVI.I. Two experimental factors were determined: concentrations with 4 levels (1%, 25%, 50% and 75%), and bioproducts with 4 levels (extracts, fractions 1, fractions 2 and fractions 3), a control factor as collection areas with 2 levels (Sincelejo and Tolú) and a variable response which was the percentage of antifungal index on mycelial growth, for a total of 160 experimental units.

RESULTS

Scientific classification of the plant material. The plant material was identified by the Herbarium of the University of Sucre as *Swinglea glutinosa*. The samples taken in the municipality of Sincelejo are stored under voucher 000073 and those collected in the municipality or Tolú with voucher 000076.

***S. glutinosa* extract from Sincelejo.** The yield of *S. glutinosa* extract collected in the municipality of Sincelejo was 4.4%, obtained as follows: $Yield\ (\%) = \left(\frac{4{,}4g}{100g}\right) X\ 100 = 4{,}4\%$

Extract of *S. glutinosa* from Santiago of the Tolú. The yield of the extract of *Swinglea glutinosa* collection in the municipality of Sincelejo was 3.8%, according to the following result: $Yield\ (\%) = \left(\frac{3{,}8g}{100g}\right) X\ 100 = 3{,}8\%$

The results obtained with respect to yield indicate higher percentages for *S. glutinosa* collected in the municipality of Sincelejo than in the municipality of Tolú. In contrast, Tolú has coastal areas with severe fertility, acidity and salinity limitations, with mangrove areas that are not suitable for agriculture (Aguilera, 2005).

Obtaining plant fractions by column chromatography. Three chromatographic fractions were obtained from each plant extract collected in the municipalities of Sincelejo and Santiago of the Tolú, for a total of 2 ethanolic extracts and 6 fractions (Figure 1).

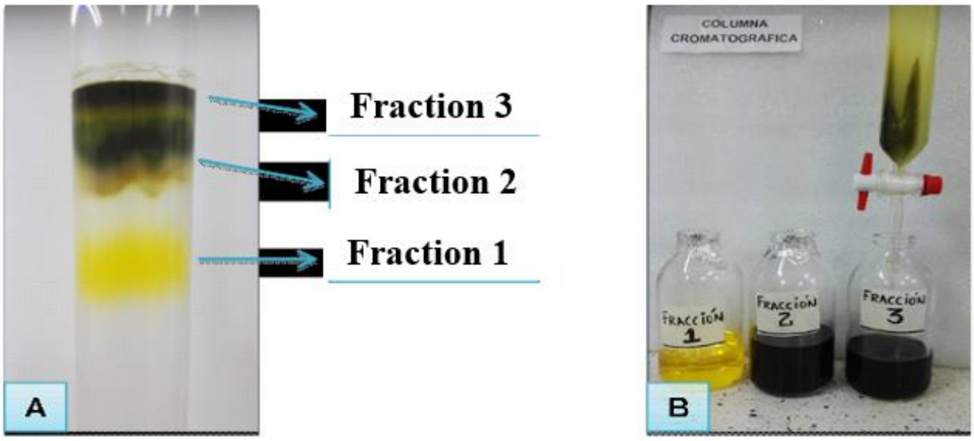

Figure 1. Obtaining the fractions from extracts of dried leaves of *S. glutinosa*.

A: chromatographic column showing the three fractions.
B: fraction 1 (Cyclohexane: Trichloroethylene (6:4)), fraction 2 (Chloroform: Acetone (8:2) and fraction 3 (Methanol).

Molecular identification of the fungus. The results of the molecular analysis performed by the CorpoGen laboratory in the city of Bogotá, the strain under study, showed an identity of 100% of its length with ITS sequences, with sequences stored in worldwide genomic banks corresponding to the species *Colletotrichum gloeosporioides* (Figure 2).

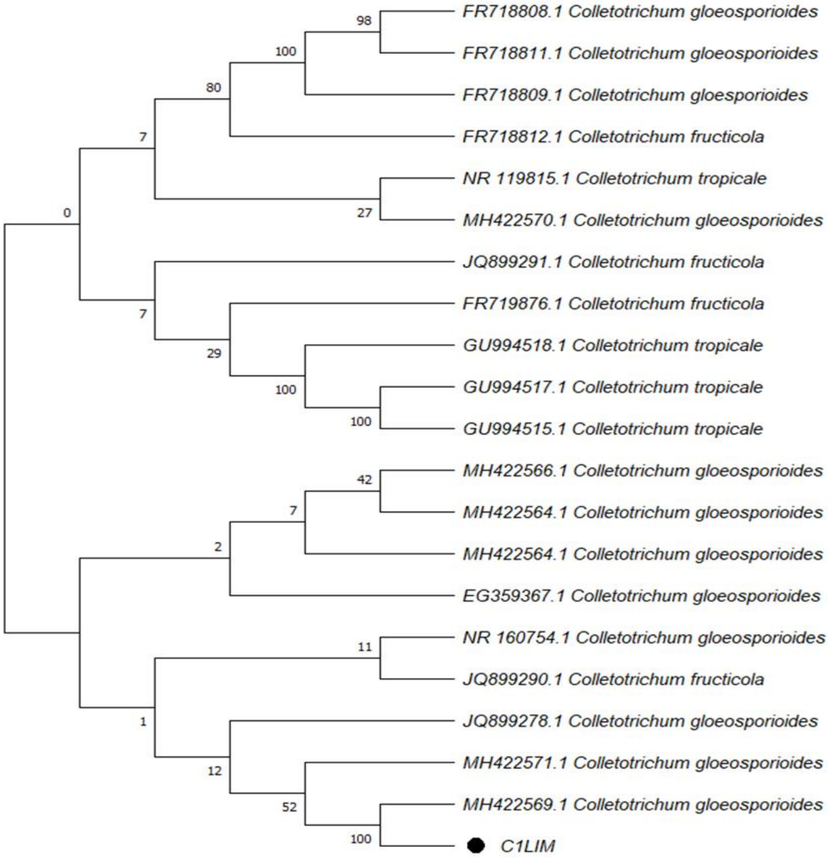

Figure 2. Distance tree constructed from the closest sequences for the fungus *C. gloesporioides*.

***In vitro* pathogenicity test of the strain *Colletotrichum gloeosporioides*.** The isolated strain of *C. gloeosporioides* was used for *in vitro* pathogenicity tests with five replicates, for which fresh leaves of the creole variety of yam (*Dioscorea alata*) were used as host, in order to guarantee pathogen-free material. The results observed after 15 days of incubation included leaf lesions typical of the disease known as anthracnose (Figure 3).

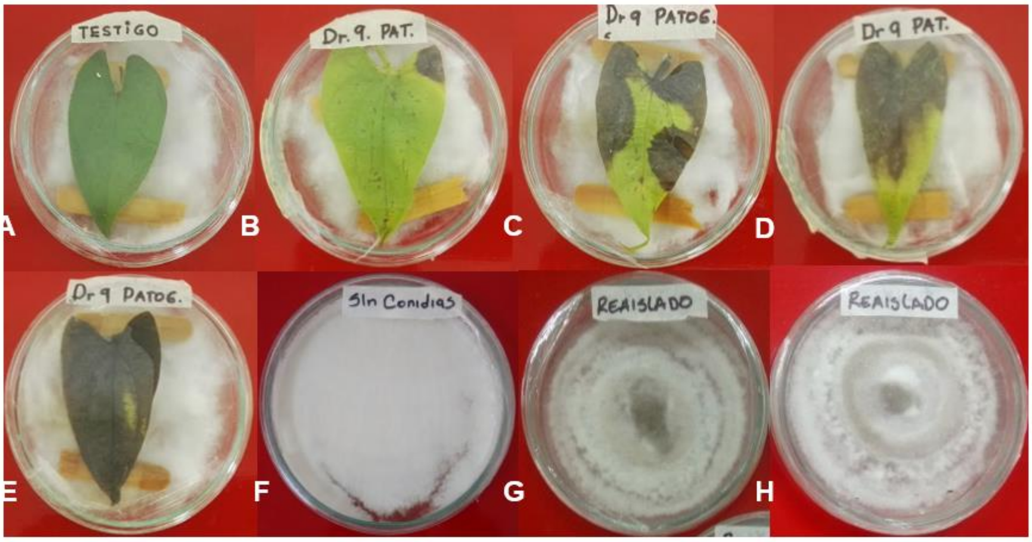

Figure 3. Pathogenicity test of *Colletotrichum gloeosporioides* on yam leaves of the Creole variety (*D. alata*). A: control, unstressed leaves. B: leaf with a 4-day inoculation strain. C: leaves with an 8-day inoculation strain. D: leaves with a 12-day inoculation strain. E: leaves with a 15-day inoculation strain. F: solution of conidia on PDA. G-H: *Colletotrichum gloeosporioides strain*

Inhibition of extracts and fractions of *Swinglea glutinosa* collected in Sincelejo against *Colletotrichum gloeosporioides*. The results obtained from the *In vitro* test of the inhibitory activity of the extracts and fractions of *S. glutinosa*, collected in the municipality of Sincelejo against the fungus *C. gloeosporioides*, indicate that the highest percentages of A.I. were observed for fraction 2 at 75% with 96.6% followed by the total extract at 75% with 87.2, the negative control had the same growth as the absolute control, indicating that the solvent used does not inhibit mycelial growth.

Inhibition of extracts and fractions of *Swinglea glutinosa* collected in Tolu against *C. gloeosporioides*. The results obtained from the *In vitro* test of the inhibitory activity of the extracts and fractions of *S. glutinosa*, collected in the municipality of Tolu against the fungus *C. gloeosporioides*, indicate that the highest percentages of A.I. were observed for fraction 2 at 75% with 76.6% followed by the total extract at 75% with 74.6%.

Minimum inhibitory concentration (MIC) and minimum fungicide concentration (MFC). The minimum inhibitory concentration (MIC) for the two ethanolic extracts of dried leaves and the two F2 fractions of *Swinglea glutinosa* was determined to be 75% and the minimum fungicide concentration (MFC) was 80%. The results obtained indicate that the minimum inhibitory concentration for both extracts and fractions 2 was 75% and the minimum fungicide was 80%, from this concentration the inhibition of mycelial growth of *C. gloeosporioides* was 100% the solvent used does not inhibit mycelial growth.

Inhibition of *Colletotrichum gloeosporioides* conidia germination. This test showed that the 25% concentration of the extracts and F2 fractions from both Sincelejo and Tolú showed the maximum inhibition (100%) of the germination of the conidia of the *Colletotrichum gloeosporioides strain*. The results obtained from the *In vitro* test of inhibition in the germination of *C. gloeosporioides* conidia indicate that the highest percentages of inhibition in the germination of conidia were observed at 100% for both extracts and fractions 2 at a concentration of 25%, similar to the positive control with Benomil at 2%. Above this concentration, the inhibition of conidia germination continued to be 100%. (Figure 4).

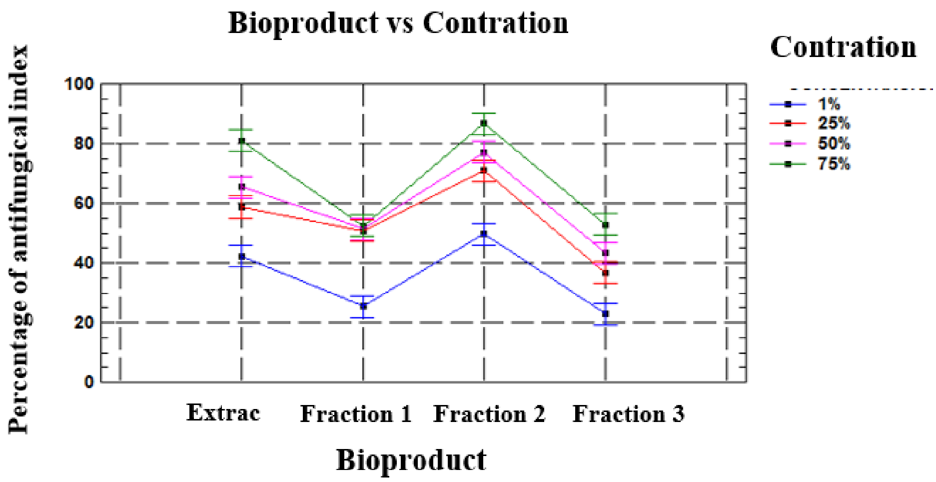

Figure 4. Interaction between the types of bioproducts and the concentrations studied on the percentage of antifungal index (%I.A).

Chemical characterization of *Swinglea glutinosa* extracts by MS-GC. The chemical analysis allowed us to establish that the antifungal activity of the extracts and fractions was not due to a particular component, since both extracts and fractions showed similarities in both the chemical compounds and their amounts. This allows us to infer that both the amount in concentration and the types of chemical species in the mixture influenced the antifungal efficacy of the extracts and fractions against Colletotrichum gloeosporioides, showing the best activities extracts and fractions 2 from Sincelejo and Tolú respectively (figure 5).

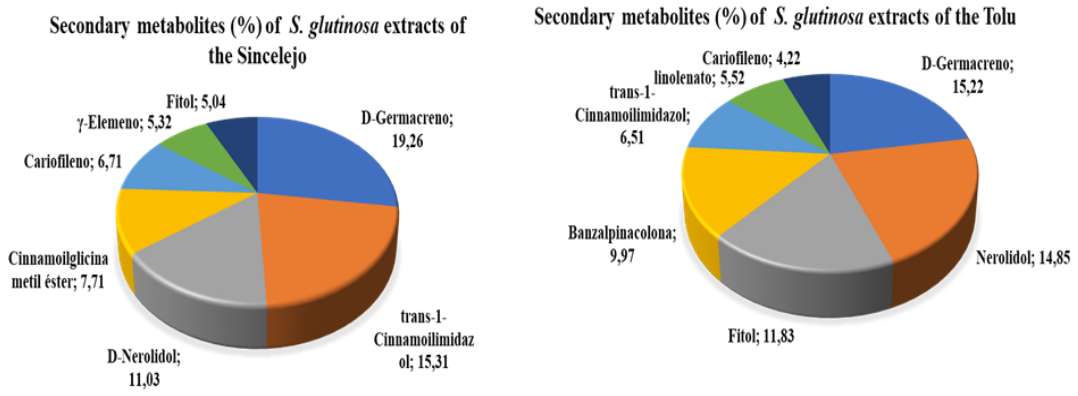

Figure 5. Chromatographic profile of the main secondary metabolites of leaf extracts *S. glutinosa* from Sincelejo and Tolú, Sucre-Colombia

DISCUSSION

It shows the interaction between the types of bioproducts and the different concentrations studied, finding that the concentrations of 75% showed the best antifungal activity against C. gloeosporioides and the bioproducts that showed the best activity were fractions 2, followed by the extracts. In general, the concentrations of the bioproducts at 75% showed the best percentage of antifungal index (%I.A), therefore, fraction 2 from Sincelejo showed the best %I.A of 96.6% at a concentration of 75%, followed by the extract from the same area and at the same concentration which was 87.2%. The antimicrobial activity of natural products is attributed to the presence of chemical compounds such as terpenes (monoterpenes and sesquiterpenes), or is attributed to alcohols and phenols, these results are similar to those found by Camargo et al. (2021) The majority components detected were β-pinene (31.3%), α-pinene (15.1%), germacrene D (14.4%), trans-nerolidol (5.6%) and sabinene (5.4%). In addition, the results maintain similarity to those of Bueno-Sanchez et al. (2009), who reported β-pinene (49.6%), α-pinene (12%) and sabinene (11.0%) as main components of the essential oil of the bark of the fruit of *S. glutinosa* of the city Bucaramanga (Colombia).

Camargo et al. (2021) performed inhibition tests of *C. gloeosporioides*, obtained from mango, and found that lemongrass extract (Swinglea glutinosa) was the most effective, in all doses used, being also effective in controlling the germination of conidia. The authors concluded that S. glutinosa essential oil exerts dose-dependent inhibition on mycelial growth and spore germination of *Colletotrichum* sp.

Regarding the work on the inhibitory activity of extracts and fractions of *Swinglea glutinosa* on the fungus *C. gloeosporioides*, which causes anthracnose disease in avocado crop, no reports were found; however, Camargo et al. (2021) found that α-pinene and β-pinene are major components (46.4%) of the essential oil *S. glutinose*, both of which have been reported for their antifungal effect. Swinglea glutinous essential oil inhibited Colletotrichum sp. depending on the concentration. A maximum inhibition of 82.41% of mycelial growth and complete inhibition of the germination of *Colletotrichum* spores in liquid medium was achieved.

Marin et al. (2008), he reported that extracts, Lemongrass (*Swinglea glutinosa*), and Lemongrass plus Neem, showed *In vitro* antifungal activity against the fungus *M. fijiensis*, and found the activity of several plant extracts with antifungal activity against pathogenic fungi of *Musa* sp. And found that *Swinglea glutinosa* presented an 80% inhibition of mycelial growth of the fungus. Likewise, Camargo et al. (2021) found that when performing the evaluation by Spore Germination Inhibition Test: The essential oil of *Swinglea glutinosa* inhibited the germination of *Colletotrichum* spores sp. by up to 100% at a concentration of 8 μL/mL. Linear regression analysis yielded an r-square of 94.23%, indicating a clear correlation between oil concentration and inhibition of conidia germination. This result shows that a considerably lower dose is required to achieve complete inhibition of germination of *Colletotrichum* sp. conidia.

The bioproducts from the two collection areas showed similarities in terms of chemical characteristics such as the presence of Carene, Benzamide, Elemene, Caryophyllene, Germacrene, among others. Although the bioproducts from Tolú showed a greater variety and quantity of chemical constituents, the best antifungal activity against *C. gloeosporioides* was shown by the bioproducts from Sincelejo. Therefore, it should be noted that the antimicrobial activity of natural products such as natural extracts depends on their chemical composition, as well as on the amount of their individual constituents (Nazarro et al., 2013). Moreover, the mechanisms of action of these natural agents are not attributed to a single mechanism but to a cascade of reactions (Burt, 2004). A possible explanation for this phenomenon is the synergism between the various components present in the mixture (Edris. 2007), since the physical properties significantly influence the action of the components, either to increase or decrease their activity (Koroch et al., 2007).

CONCLUSIONS

This study evaluated the inhibitory effect of extracts and fractions obtained from *Swinglea glutinosa* on a strain of *Colletotrichum gloeosporioides* fungus causing anthracnose disease of avocado in the department of Sucre-Colombia. The results obtained from the inhibitory activity show that at high concentrations a higher antifungal index (%I.A) was observed for the bioproduct fraction 2 and the Sincelejo extract with a %I.A of 96.6% and 87.2% respectively, which makes this *In vitro* study a potential alternative in the future for the field management of anthracnose caused by Colletotrichum species in the avocado crop in the department of Sucre. It was determined that the minimum fungicide concentration was 80% for the extracts and fractions 2 of the two municipalities, similar activity shown by the positive control with Benomyl at 2%. In addition, both extracts and fractions 2 showed 100% inhibition of germination of *C. gloeosporioides conidia* at 25% concentration. The extract and fractions of *Swinglea glutinosa* have several characteristics that make it important for the current agricultural activity; on the one hand, it is beneficial for the environment and for the people who apply it; on the other hand, the cost of processing at the farm level is significantly lower than chemicals that perform the same function.

ACKNOWLEDGEMENTS

To the Colombia Científica Program "Reconstruction of the social fabric in post-conflict zones in Colombia" PGIS Code: 57579 with the research project "Entrepreneurial and innovation competencies for economic development and productive inclusion of regions affected by the Colombian conflict" PGIS Code: 58907. Funded within the framework of the call Scientist Colombia, Contract No FP44842-213-2018.

AUTHOR CONTRIBUTION. Alexander Perez Cordero: experiment execution, data analysis. Donicer Montes V and Pavel Peroza-Piñere, conceptualization, writing - revision and editing. All authors have read and approved the manuscript.

CONFLICT OF INTEREST. The authors of the manuscript, declare that there is no conflict of interest related to the article.

REFERENCES

1. Bueno-Sánchez J. G., Martínez-Morales J. R., Stashenko E. E., & Ribón, W. (2009): Anti-tubercular activity of eleven aromatic and medicinal plants occurring in Colombia. Biomedica, 29: 51-60.
2. Camargo Piñeres Y., Zambrano Montenegro G., Ortega-Cuadros M., Gutierrez Montero, D. J., & Yepes, J. A. (2021): Actividad antifúngica *in vitro* del aceite esencial de *Swinglea glutinosa* Merr sobre *Colletotrichum* sp. patógeno de mango (*Mangifera indica* L). Revista Colombiana de Biotecnología, 23: 62-71.

3. García-Mateos, M. R., Acosta-Ramos, M., Rodríguez-Pérez, E., Vásquez-Sánchez, J., & Hernández-Ramos, L. (2021). Extractos vegetales para el control de Colletotrichum gloeosporioides in vitro, en periodo de floración y poscosecha del fruto de Carica papaya. Polibotanica, 51: 213-228.
4. Alarcón R. J., Chavarriaga M. (2007): Diagnóstico precoz de la antracnosis (*Colletotrichum gloeosporioides)* (Penz) Penz and Sacc. En tomate de árbol mediante el empleo de infecciones quiescentes. Agron, 15: 89-102.
5. Burt, S. (2004): Essential oils: their antibacterial properties and potential applications in foods-A review. Int. J. Food Microbiol, 94: 223–253.
6. Burt S.A., Van Der Zee, R., Koets, A.P., de Graaff, Van Knapen, F., Gaastra, W., Haagsman, H.P., Veldhuizen, E.J. (2007): Carvacrol induces heat shock protein and inhibits synthesis of flagellin in *Escherichia coli* O157:H7. Appl. Environ. Microbiol, 73: 4484–4490.
7. Consentin S., Tuberoso,C.I., Pisano B., Satta M., Mascia V., Arzedi E., Palmas F. (1994): *In-vitro* antimicrobial activity and chemical composition of *Sardinian Thymus* essential oils. Lett Appl Microbio, 29:130-135.
8. Cowan M. (1999): Plant products as antimicrobial agents. clin. microbiol. rev. 10: 564-582.
9. Edris A.E. (2007): Pharmaceutical and therapeutic potentials of essential oils and their individual volatile constituents: a review. Phytother, 21:308-323.
10. Espitia-Baena J.E., Robledo-Restrepo S.M., Cuadrado-Cano B.S., Duran-Sandoval H.R., Gómez-Estrada H.A. (2014): Perfil fitoquímico, actividad anti-*Leishmania*, hemolítica y toxicológica de *Cordia dentata* Poir. y *Heliotropium indicum* L. Revista Cubana de Plantas Medicinales, 9: 1-21.
11. Stashenko E., Martínez J. R., Medina J. D., & Durán C. (2015): Analysis of essential oils isolated by steam distillation from Swinglea glutinosa fruits and leaves. Journal of Essential Oil Research, 27: 276-282.
12. Gil A. I., Celis Á., & Cuevas J. C. (2010): Efecto inhibitorio de extractos de *Swinglea glutinosa* (Blanco) Merr. y Lantana camara L. en preemergencia y posemergencia. Revista Colombiana de Ciencias Hortícolas, 4: 223-234.
13. Hincapié O. D., Saldarriaga A., Díaz, C. (2017): Biological, botanical and chemical alternatives for the control of blackberry (*Rubus glaucus* Benth.) diseases. Revista Facultad Nacional de Agronomía, 70: 8169- 8176.
14. Hurtado E., Fernández A., Carrasco A. (2018): Avocado fruit *Persea americana*. (eds.): Amsterdam, Netherland: Elsevier, 37–48.
15. Koroch, A. R., Rodolfo Juliani, H., & Zygadlo, J. A. (2007): Bioactivity of essential oils and their components. Flavours and fragrances, 87-115.
16. Landero-Valenzuela N., Lara-Viveros F.M., Andrade-Hoyos P., Aguilar-Pérez L.A., Aguado, G.J. (2016): Alternativas para el control de *Colletotrichum* spp. Revista Mexicana de Ciencias Agrícolas, 7: 1189- 1198.
17. Macias F. A., Castellano D., Oliva R. M., Cross P., Torres A. (1997): Potential use of allelopathic agents as natural agrochemicals. (eds.): Elsevier: 37–48.
18. Marin O. J., M.J Barrera., J Robles., J. (2008): Evaluación de extractos vegetales para el control de *Mycosphaerella fijiensis* en platano en Tierralta- Córdoba. Temas Agrarios, 1: 25-31.
19. Nazarro F., Fratianni F., De Martino L., Coppola R., De Feo V. (2013): Effect of Essential Oils on Pathogenic Bacteria. Pharmaceuticals, 6: 1451-1474.
20. Torres-González C., Casas M., Díaz Ortiz J. E. (2013): Manejo de *Ralstonia Solanacearum* raza 2 a través de productos químicos y biológicos. Iteckne, 10: 217-223.
21. Ramírez J. G., Gonzalo J., Peterson A. (2018): Potential geography and productivity of "Hass" avocado crops in Colombia estimated by ecological niche modeling. Scientia Horticulturae. 237: 287–295.
22. Rao G., Singh M., OSingh H. (1992): Fungitoxic evaluation of essential oils extracted from higher plants against some sugarcane pathogens *in vitro*. Tropic Sci, 32: 377-382.

23. Rimá de Oliveira K. Á., Ramos Berger L. R., Amorim de Araújo S., Saraiva Câmara M. P., & Leite de Souza, E. (2017): Synergistic mixtures of chitosan and *Mentha piperita* L. essential oil to inhibit *Colletotrichum* species and anthracnose development in mango cultivar Tommy Atkins. Food Microbiology, 66: 96-103.
24. Rondón O., Sanabría de Albarracín N., Rondón, A. (2008): Respuesta *In vitro* a la acción de fungicidas para el control de antracnosis, *Colletotrichum gloeosporioides* Penz, en frutos de mango. Agronomía Tropical, 56: 219-235.
25. Cifuentes-Arenas J. C., Beattie G. A. C., Peña L., Lopes, S. A. (2019): Murraya paniculata and Swinglea glutinosa as short-term transient hosts of 'Candidatus Liberibacter asiaticus' and implications for the spread of huanglongbing. Phytopathology, 109(12): 2064-2073.
26. Pérez-Cordero A., Chamorro-Anaya L., Mercado-Gómez J. D. (2021): Inhibición de *Colletotrichum gloeosporioides* en cultivos de ñame en el Caribe colombiano usando aceites esenciales de Curcuma longa y Zingiber officinale. Ciencia en Desarrollo, 12: 1-12.