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In vitro antifungal activity of *Lippia graveolens* and *Viscum album* ethanolic extracts against *Fusarium oxys*porum f. sp. *lycopersici*

Actividad antifúngica in vitro de extractos etanólicos de *Lippia graveolens* y *Viscum album* contra *Fusarium oxysporum* f. sp. *lycopersici*

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Technological Innovation: Evaluation of the antifungal activity of two plant extracts against the phytopathogenic fungus *Fusarium oxysporum* f. sp. *lycopersici*.

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Resumen

La marchitez vascular causada por *Fusarium oxysporum* f. sp. *lycopersici* es un problema en el cultivo de tomate. Su manejo comúnmente se hace con productos sintéticos. Sin embargo, el uso de este tipo de productos podría inducir resistencia hacia los ingredientes activos y problemas toxicológicos. Debido a esto, se han propuesto alternativas bioracionales como los extractos vegetales, para el manejo de enfermedades en las plantas. Los objetivos de esta investigación fueron obtener información de la composición fitoquímica de los extractos vegetales, así como determinar la actividad antifúngica y la concentración inhibitoria al 50 y 90 % (CI50 y CI90) de extractos vegetales etanólicos de *Lippia graveolens* y *Viscum album* mediante el método de microdilución en placa; se realizó un análisis Probit para determinar la CI50 y CI90 de cada extracto, y ANOVA por prueba de Tukey ($p<0.05$) para determinar diferencias estadísticas. En ambos extractos se identificaron compuestos fitoquímicos, como

ácidos hidroxicinámicos, estilbenos, flavonoides, catequinas, polifenoles y ácidos metoxicinámicos. Además, ambos extractos presentaron actividad antifúngica mayor al 80 % de inhibición. Sin embargo, el extracto de *L. graveolens* fue estadísticamente mejor, con una CI_{50} más baja de 60.91 mg/L. En conclusión, ambos extractos vegetales contienen compuestos fitoquímicos de interés para el control de microorganismos y representan una alternativa natural para el control de *F. oxysporum* f. sp. l*ycopersici*.

Palabras clave: Fungicidas botánicos, Extractos de plantas, Compuestos fitoquímicos, Control biológico de enfermedades, Biofungicidas.

Abstract

Vascular wilt caused by *Fusarium oxysporum* f. sp. *lycopersici* is a problem in tomato crops. Its management is commonly done with synthetic products. However, the excessive use of such products could induce resistance to the active ingredients and toxicological problems. Because of this, biorational alternatives such as plant extracts have been proposed for plant disease management. The objectives of this research were to obtain information on the phytochemical composition of plant extracts, as well as to determine the antifungal activity and the inhibitory concentration at 50 and 90 % (IC50 and IC90) of ethanolic plant extracts of *Lippia graveolens* and *Viscum album* using the microdilution plate method. A Probit analysis was performed to determine the IC50 and IC90 of each extract, and ANOVA by Tukey's test ($p<0.05$) to determine statistical differences. Phytochemical compounds, such as hydroxycinnamic acids, stilbenes, flavonoids, catechins, polyphenols, and methoxycinnamic acids were identified in both extracts. Furthermore, both extracts showed antifungal activity higher than 80 % inhibition. However, the *L. graveolens* extract was statistically superior, with the lowest IC50 of 60.91 mg/L. In conclusion, both plant extracts contain phytochemical compounds of interest for the control of microorganism and represent a natural alternative for controlling *F. oxysporum* f. sp. *lycopersici.*

Keywords: Botanical fungicides, Plant extracts, Phytochemical compounds, Biological control of diseases, Biofungicides.

1. Introduction

Tomato farming is one of the world´s major crops, being the second most cultivated vegetable, second only to potatoes, with an estimated production of 140 million tons (FAO, 2022). Nevertheless, the production and quality of tomatoes are affected by several diseases caused by viruses, bacteria, oomycetes, and fungi in fields, greenhouses, on seedlings, and at the post-harvest stage (Singh *et al*., 2017). Vascular wilt by *F. oxysporum* f. sp. *lycopersici* is one of the main diseases in tomato-producing areas, which, if not controlled, can cause losses in production of up to 80 % because it affects the vascular system of plants, causing wilt (Marlatt *et al.*, 1996; González *et al.*, 2012; Hernández- Martínez *et al.*, 2014). Currently, the control of this disease is with chemical products; however, the continues use of these products has caused resistance on microorganisms toward the active ingredients and ecological problems too (Chen *et al.*, 2007).

Given the aforementioned challenges, the demand for organic products worldwide is on the rise, reflecting a global shift towards sustainable agriculture. In response, researchers are actively exploring new

compounds for disease control that do not compromise environmental security (Hernández *et al*., 2007; Igbinosa *et al*., 2009). This has led to the emergence of plant extracts with antifungal properties as a promising alternative for plant disease management. Numerous studies have demonstrated their efficacy in controlling phytopathogens, thanks to the diverse array of secondary metabolites they contain, such as flavonoids, phenols, terpenes, essential oils, alkaloids, lectins, and polypeptides, each with its unique mode of action (Castillo *et al*., 2010; Mendez *et al*., 2012).

The vegetable species *L. graveolens* and *V. album* are options to produce plant extracts with antimicrobial activity; in the case of *L. graveolens*, it has a well-documented antifungal capacity, inhibiting the development of phytopathogenic fungi such as *Rhizopus stolonifer*, *Colletotrichum gloesporoides*, *Penicillium digitatum*, and *Rhizoctonia solani* (Hernández-Castillo *et al*., 2010; De Rodríguez *et al.*, 2011), becoming an alternative for plant extracts production for disease management; while *V. album* is a species that has been reported to possess antimicrobial activity and induce resistance in plants towards phytopathogenic microorganisms due to the considerable amount of phytochemical compounds present in its metabolism, becoming a viable option to be used in disease management on crops (Chandrashekhara *et al.*, 2010; Orhan *et al.*, 2010; Hussain *et al.*, 2011; Nacsa-Farkas, 2014). Both plant extracts contain many phytochemical compounds, such as polyphenols, tannins, stilbenes, flavonoids, and saponins. These compounds can affect the development of phytopathogenic fungi through various mechanisms of action, such as inhibiting electron transport and oxidative phosphorylation, damaging DNA, inhibiting key proteins for cell division, and inhibiting genes involved in the production of ergosterol in fungal cell walls (Jasso de Rodriguez *et al*., 2011; Mendez *et al*., 2012; Cantelli *et al.*, 2017; Vestergaard & Ingmer, 2019).

The aims of this research were to obtain information about the phytochemical composition of plant extracts from *L. graveolens* and *V. album*, and determine its biological effectiveness in *in vitro* tests of these extracts against *F. oxysporum* f. sp. *lycopersici* isolated from diseased tomato plants.

2. Materials and methods

Collection of plant species

The sampling of *L. graveolens* was performed in the municipality of General Cepeda, Coahuila, Mexico; the plant specimens were placed into plastic bags and then transported to the laboratory of Mycology and Biotechnology of the Autonomous University Agrarian Antonio Narro; while the *V. album* plants were collected in the municipality of Orizaba, Veracruz, Mexico. Both plant species were in the flowering phenological stage in august 2023. Whole plant samples were washed and cut into pieces of 1 cm; subsequently, the samples were placed in a drying stove at 60 °C until they showed constant weight; finally, the plants were pulverized, sifted with a pore size of 0.2 mm, and stored in a flask in the darkness at room temperature (Castillo *et al.*, 2010).

Preparation of Plant extracts

Plant extracts were produced using the method proposed by Shami *et al.* (2013). The solvent was ethanol, and the initial step involved adding 14 g of pulverized plant material in 200 mL of solvent and placing it under agitation (LabTech, LMS-1003, Korea) for 72 h at 50 °C (Jasso de Rodríguez *et al.*, 2015). Subsequently, the obtained extract was filtered with Wathman No. 1 paper to separate the extract from the dust; the extract was roto-evaporated (Yamato, RE301B-O, Japan) at 150 RPM at 60 °C; it was lyophilized and finally stored at – 20 °C.

Identification of phytochemical compounds by analytical High-Performance Liquid Chromatography-Electrospray Ionization-Mass Spectrometry (HPLC-ESI-MS)

HPLC-ESI-MS was employed to identify the phytochemical compounds, following the protocol described by Araújo *et al*. (2020). The chromatography setup included a Varian HPLC system, with an autosampler (Varian ProStar 410, USA), a ternary pump (Varian ProStar 230I, USA), and a PDA detector (Varian ProStar 330, USA); and ion trap liquid chromatography mass spectrometer equipped with an electrospray ion source was also used. Five microliters of the sample were injected into Denali C18 column (150 mm × 2,1 mm, 3µm, Grace, EE. UU.); the temperature of the oven was maintained at 30 °C, the used eluents were formic acid (0.2 %, v/v; solvent A) and acetonitrile (solvent B). The applied gradient was, initially, 3 % B; 0–5 min, 9 % B linear; 5–15 min, 16 % B linear; 15–45 min, 50 % B linear; then, the column was washed and reconditioned. The flow rate was maintained at 0.2 mL/min, and the elution was monitored at 245, 280, 320, and 550 nm; the whole effluent was injected into the spectrometer source without splitting. All MS experiments were carried out in negative [M-H]-1mode. Nitrogen was used as nebulizing gas and helium as damping gas; the parameters of the ion source were: spray voltage of 5.0 kV and capillary voltage of 90.0 V, as well as the temperature of 350 °C. The data were analyzed with the software MS Workstation (V6.9), and the samples were firstly analyzed in full scan mode acquired in the m/z range 50–2000.

Determination of total phenols, flavonoids, and antioxidant activity by DPPH

The total phenolic content was determined in both extracts, following the methodology proposed by Zenil-Lugo et al. (2014) with slight modifications. The extracts were resuspended at a concentration of 1000 mg/L using water as solvent. Subsequently, 20 µL of the extract, control, and calibration curve (98, 195, 391, 781, 1563, 3125 mg/L, r2= 0.998) were mixed with 120 µL of Na2CO3, 30 µL of Folin-Ciocalteu reagent, and 400 µL of water. The reaction was conducted at 50 °C for 5 minutes, and absorbance values were measured at 700 nm using a microplate reader (Thermo Scientific, Multiskan GO, Finland). Gallic acid was used as the standard reference. Total flavonoids were determined using the aluminum chloride method described by Chang *et al*. (2002), with absorbance measured at 510 nm. Quercetin was used as standard reference, and a calibration curve was prepared using quercetin solutions (195, 391, 781, 1563, 3125, 6250, 12500, 25000, 50000, 100000 mg/L, r2= 0.998). Antioxidant activity was assessed following the method outlined by Jasso de Rodríguez *et al*. (2015). A 1-diphenyl-2-picrylhydrazyl (DPPH) solution was used, with 2950 µL of the solution added to 50 µL of the extract. The mixture was stirred manually and incubated for 30 minutes at 37 °C, and absorbance was subsequently measured at 517 nm. A control solution consisting of 100 µL of distilled water was used for comparison. The results were expressed as the percentage of inhibition, calculated using the following formula:

$$\%\ of\ inhibition\ DPPH = \frac{1 - Abs\ 517nm\ sample}{1 - Abs\ 517nm\ control} x\ 100$$

Biological effectiveness of plant extracts of *L. graveolens* and *V. album* in *vitro*

Obtaining F. *oxysporum* f. sp. *lycopersici* strain and molecular identification

F. oxysporum f. sp. *lycopersici* strain was isolated from tomato plants showing symptoms of vascular wilt, collected from the tomato production region of the municipality of Ahome, Sinaloa, Mexico. Previously a pathogenicity test was conducted with this strain in tomato seedlings, resulting in wilt within seven days. The DNA extraction was done using the CTAB method according to Kumar *et*

al. (2014), with some modifications, employing the previously isolated strain cultured on Potato Dextrose Agar medium for five days. PCR was performed using the primers FO1 (5′-ACATACCACTTGTTGCCTCG-3′) and FO2 (5′ CGCCAATCAATTTGAGGAACG-3′) (Nirmaladevi *et al.*, 2016). The amplification mixture consisted of 3 µL of DNA from the sample, 12.5 µL of GoTaq® Green Master Mix (GoTaq® DNA polymerase in 2X reaction buffer GoTaq® pH 8.5, 400 µg dNTPs, 3 mM MgCl2, reaction buffer), 2 µL of primers and 7.5 µL of sterile water nuclease-free. The conditions of amplification were initial denaturalization at 94 °C for 4 min, followed by 30 cycles at 94 °C for 1 min, 58 °C for 1 min, and 72 °C for 1.5 min, with a final extension of 72 °C for 8 min. The product of PCR was visualized on agarose gel at 1 % with ethidium bromide and visualized with light UV (Nirmaladevi *et al.*, 2016). The isolated and identified strain was added to the strain collection of the Laboratory of Mycology and Biotechnology within the Parasitology Department of the Autonomous University Agrarian Antonio Narro.

Antifungal activity of plant extracts against *F. oxysporum* f. sp. *lycopersici* by the micro-dilution in plate method.

The microplate dilution method used was that described by Tucuch-Pérez *et al.* (2020) with slight modifications; the treatments were the plant extracts of *L. graveolens* and *V. album*, which were resuspended at 2000 mg/L using water as solvent. Polystyrene 96- well plates were filled with 100 µL of Sabouraud liquid medium; after placing 100 µL of each extract at 2000 mg/L in column number four it was mixed, and 100 µL of the mixture was transferred to column number five, the same action was carried out up to column number 12, obtaining concentrations of 1000 mg/L, 500 mg/L, 250 mg/L, 125 mg/L, 62.5 mg/L, 31.2 mg/L, 15.6 mg/L, 7.8 mg/L y 3.9 mg/L (figure 1); the next step was to add 40 µL of 2,3,5-triphenyl tetrazolium chloride; finally 10 µL of a conidia solution at $1x10^8$ was added in all wells of plate except in the wells of column number one. Each plate was considered a repetition and eight repetitions per treatment were perfomed; the plates were incubated at 28 °C, and with a spectrophotometer, the absorbance of plates was read at 490 nm (Thermo Scientific, Multiskan GO, Finland). The growth of the fungus was positive when the well-showed pink coloration and negative when no coloration was present. The inhibition percentage was calculated by considering it as the inverse of the growth percentage, using the formulas:

$$\%\ \text{of growth} = \frac{(A - B)}{C} x\ 100$$

Where A is the absorbance of each treatment, B is the absorbance of the negative control, and C is the absorbance of the positive control.

$$\%\ \text{of inhibition} = 100 - \%\ of\ growth$$

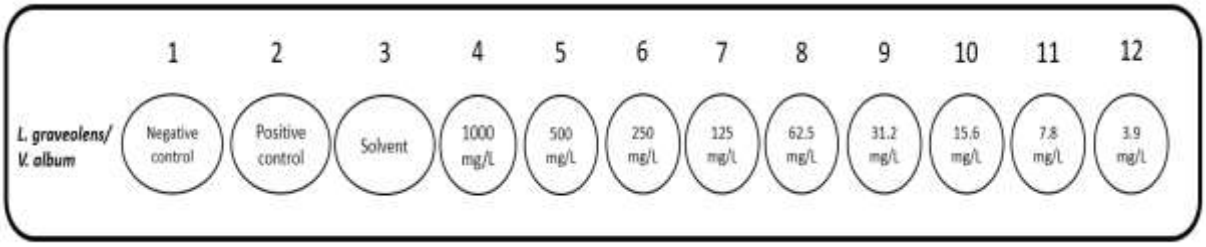

Figure 1. Arrangement of treatments and concentrations used in the in vitro bioassay on 96-well polystyrene plates.

Statistically analysis

A Probit analysis was carried out for both bioassays to determine the IC50 and IC90 of each extract; subsequently, An ANOVA was performed with the results obtained using the Tukey's test ($p<0.05$) with the Statistical Analysis System (SAS) 9.0 software.

3. Results and discussion

Identification of phytochemical compounds by analytical HPLC-ESI-MS

Table 1 shows the compounds detected in *L. graveolens* and *V. album* extracts by HPLC-ESI-MS analysis, where the presence of compounds from the hydroxycinnamic acids, stilbenes, flavonols, methoxy flavones, catechins, polyphenols, and flavanones families can be observed in the extract of *L. graveolens*, and compounds from hydroxycinnamic acids, catechins, flavones, and methoxycinnamic acid families were observed in the extract of *V. album*. Regarding this, Trejo-Márquez *et al.* (2015) reported the presence of resveratrol and caffeic acid in an ethanolic extract from *L. graveolens*. Furthermore, the presence of caffeic acid in *Lippia citriodora* extract has been reported (Quirantes-Piné *et al.*, 2009); on the other hand, chromatographic studies by HPLC have detected the presence of caffeic acid in methanolic extracts of *V. album* (Khatun *et al.*, 2016). The antioxidant activity, which allows the interaction of phytochemical compounds with free radicals, is one of the most significant attributes of flavonoids because it is related to the antifungal activity by the interaction with fungal reactive oxygen species (Warris & Ballou, 2019), kaempferol and catechins have been identified as common compounds present in plant extracts produced from various solvents of species within the Verbenaceae family, which includes the genus *Lippia* (Güereca *et al.*, 2007; Funari *et al.*, 2012; de Oliveira *et al.*, 2013; Cid-Pérez *et al.*, 2016), while in ethanolic extracts from *V. album* it has been possible to detect catechins (Kang *et al.*, 2016; Trifunschi *et al.*, 2017). Among the secondary metabolites of plants, the polyphenols play an essential role in the antimicrobial and antioxidant activity present in plant extracts; this capacity to inhibit the growth of the microorganisms has been documented in several studies, allowing to obtain natural compounds as alternatives to control diseases in plants such as the arbutin, pinocembrin, luteolin, and ferulic acid (López-Lázaro, 2009; Rasul *et al.*, 2013; Jurica *et al.*, 2017; Kim & Park, 2019). These compounds, as mentioned earlier, have been reported by several authors as compounds present in plant extracts; in this sense Arias *et al.* (2020) detected pinocembrin in the essential oil of *Lippia origanoides* and *L. graveolens*, whereas in extracts from *V. album* the presence of luteolin and feruloylquinic acid has been detected (Trifunschi *et al.*, 2017; Stefanucci *et al.*, 2020).

Table 1. Detection of phytochemical compounds in extracts from *L. graveolens* and *V. album* by HPLC-ESI-MS.

Extract	Compound	Retention time (Min)	Mass	Family
L. graveolens	Caffeic acid 4-O-glucoside	4.094	340.9	Hydroxycinnamic acids
	Resveratrol 3-O-glucoside	13.881	391	Stilbenes
	Resveratrol 5-O-glucoside	18.291	389	Stilbenes
	Kaempferol 3-O-glucosyl-rhamnosyl-glucoside	25.897	755	Flavonols
	Sinensetin	26.243	371.1	Methoxyflavones
	(+)-Catechin 3-O-glucose	26.699	451.3	Catechins

	Arbutin	41.333	270.9	Other polyphenols
	Pinocembrin	50.559	254.9	Flavanones
V. album	Caffeic acid 4-O-glucoside	4.152	341	Hydroxycinnamic acids
	(+)-Catechin 3-O-glucose	26.628	451.3	Catechins
	Luteolin 7-O-(2-apiosyl-6-malonyl)-glucoside	3.823	665	Flavones
	5-Feruloylquinic acid	55.776	367.2	Methoxycinnamic acids
	p-Coumaroyl tartaric acid	58.2	295.2	Hydroxycinnamic acids

Determination of total phenols, total flavonoids, and antioxidant activity by DPPH

Statistical differences were observed among the extracts of *L. graveolens* and *V. album* in determining total phenols and flavonoids. However, no statistical differences were observed in the evaluation of antioxidant activity (Table 2). Phenols and flavonoids are synthesized in all plant organisms and exhibit high biological activity when applied to different organisms. Previous studies have analyzed the presence of total phenols, flavonoids, and antioxidant activity in *L. graveolens* extracts, reporting 4.54 mg/g total phenols, 200 mg/g total flavonoids, and an antioxidant activity of 8.79 % (Martínez-Rocha *et al.*, 2008; Cortés-Chitala *et al.* 2021). Additionally, other studies reported 10.43 mg/g and 0.428 mg/g of total phenols and flavonoids respectively in extracts from *V. album* (Pietrzak *et al.*, 2017) and an antioxidant activity of 95.36 % (ÖnayUçar *et al.*, 2006).

Table 2. Total phenols and flavonoids, and antioxidant activity (DPPH) of ethanolic extracts from *L. graveolens* and *V. album.*

Extract	Total phenols (mg/g)*	Total flavonoids (mg/g)	DPPH (%)
V. album	53±17b	283±85b	94±0.24a
L. graveolens	254±3a	1210±349a	96±0.39a

* = Values with the same letter are statistically equal (P≤0.005).

Biological effectiveness of plant extracts of *L. graveolens* and *V. album* in *vitro*

Molecular identification of *F. oxysporum* f. sp. *lycopersici* strain

The amplicon obtained with the specific primers FO1/FO2 confirmed the species *F. oxysporum* f. sp. *lycopersici* obtaining 340 bp amplified DNA products (Nirmaladevi *et al.*, 2016).

Antifungal activity of plant extracts against *F. oxysporum* f. sp. *lycopersici* by micro-dilution plate method.

The bioassays using the microdilution plate method demonstrated significant inhibition of the pathogen by *L. graveolens* and *V. album* extracts. The inhibitory effect of *L. graveolens* extract on *F. oxysporum* f. sp. *lycopersici* was observed starting at a concentration of 62.5 mg/L, with an inhibition rate of 41.72 %. Subsequently, as the concentration increased to 125 mg/L, 250 mg/L, and 1000 mg/L, the inhibition rates progressively increased to 50.83 %, 72.08 %, 83.61 %, and eventually 88.65 %. On the other hand, the inhibitory activity of *V. album* extract started at 125 mg/L, showing an inhibition rate of 50.25 %. As the concentration increased to 500 mg/L and 1000 mg/L, the inhibition rates increased to 73.75 % and 88.56 %, respectively. These results showed the inhibitory activity of both extracts against *F. oxysporum* f. sp. *lycopersici*, with their efficacy being concentration-dependent. (Figure 2).

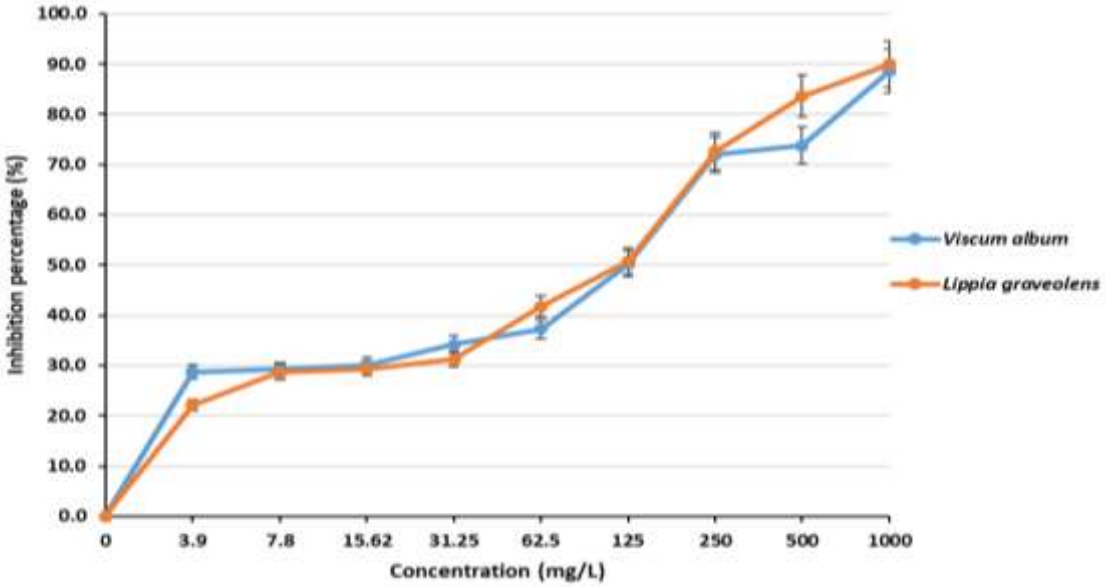

Figure 2. Inhibition percentage of ethanol extracts from *Lippia graveolens* and *Viscum album* on *Fusarium oxysporum* f. sp. *lycopersici* by microdilution in plate method.

The antimicrobial activity from *L. graveolens* and *V. album* extracts can be due to resveratrol, a stilbene detected in plant extracts. Studies have reported that it acts on ATP hydrolysis, inhibits oxidative phosphorylation, induces DNA fragmentation, suppresses the expression of ftsZ protein, and is correlated with cell membrane damage (Vestergaard & Ingmer, 2019). Flavonoids are compounds recognized for their antioxidant activity. Recent studies have shown that flavonoids could inhibit hyphae formation because they can affect reactive oxygen species of pathogen's mitochondria (Candiracci *et al.* 2012; Warris & Ballou 2019). The luteolin inhibits the efflux pump, and pinocembrin decreases ATP production, causing lipid peroxidation (Peng *et al.*, 2012). On the other hand, caffeic acid and feruloylquinic acid inhibit gene expression involved in ergosterol synthesis (Cantelli *et al.*, 2017). Finally, catechins and saponins damage the cell membrane, causing lysis of hyphae and forming a complex with sterols, allowing higher permeability of the fungal membrane (Hirasawa & Takada, 2004; Lanzotti *et al.*, 2012).

Several studies have demonstrated the antifungal activity of *L. graveolens* extract. Medina-Romero *et al*. (2022) reported inhibition of radial growth, damage to hyphal morphology, and effects on spore production and germination of *Fusarium* species treated with pure carvacrol and the essential oil of *L. graveolens*. Additionally, De Rodríguez *et al*. (2011) reported a 100 % inhibitory effect on mycelial and sporulation growth of *C. gloesporoides* and *R. stolonifer* using an ethanolic extract of *L. graveolens* at 500 mg/L, while Hernández-Castillo *et al*. (2010) used ethanol to produce an extract of *L. graveolens* and reported a maximum mycelial inhibition of 60 % against *R. solani* at 3000 mg/L. On the other hand, Ertürk *et al*. (2004) and Hussain *et al*. (2011) observed antifungal activity by testing ethanolic and hexane extracts from *V. album* against *Candida albicans*, *Saccharomyces cerevisiae*, and *Aspergillus flavus*. Ertürk (2010) reported inhibition by the ethanolic extract of *V. album* on *C. albicans* and *Aspergillus niger*. The antifungal activity of both extracts might be attributed to the various modes of action of compounds identified therein.

IC50 of *L. graveolens* and *V. album* extracts

The IC50 values of the extracts against *F. oxysporum* f. sp. *lycopersici* exhibited

statistically significant differences, as shown in Table 3.

Table 3. Inhibitory concentration to 50 % and 90 % (IC50 and IC90) of ethanolic extracts from *L. graveolens* and *V. album* on *F. oxysporum* f. sp. *lycopersici*.

Source of plant extract	IC50* (mg/L)	IC90 (mg/L)
L. graveolens	60.91 ± 0.07a	1990 ± 0.07a
V. album	63.73 ± 0.09b	3892 ± 0.09b

*=Values with the same letter are statistically equal (P≤0.005).

The IC50 indicates the amount of a compound necessary to inhibit a biological process by half, allowing for the measure of inhibitory capacity. The IC50 is crucial because a low IC50 of an agricultural product positions it as an excellent candidate for controlling phytopathogens (Aykul & Martinez-Hackert, 2016). Tests were conducted to evaluate the inhibitory effect of plant extracts by determining their IC50 values. Regarding the plants investigated in our study, Hernández-Castillo *et al*. (2010) reported an IC50 of 1930 mg/L for the ethanolic extract of *L. graveolens* against *R. solani*. However, there are no documented reports of *V. album* extract has been tested against phytopathogenic microorganisms, as compounds derived from this species commonly treat human diseases.

Nevertheless, it is noteworthy that ethanolic extracts of *V. album* have been reported to exhibit a low IC50 value in destroying carcinogenic cells (Zhao *et al*., 2012). The IC50 obtained in this work for *F. oxysporum* f. sp. *lycopersici* with both plant extracts can be considered low due to the presence of phytochemical compounds, such as polyphenolic compounds, which inhibit the development of phytopathogenic microorganisms. These compounds act by the interaction of phenolic hydroxyl groups with the hydrophilic ends of the lipid bilayer to agglomerate lipids of the membrane and provoke its rupture, allowing the leakage of cytoplasmic material (Yang & Zhang, 2019).

4. Conclusions

L. graveolens and *V. album* are plant species widely used in plant extract production, with their application in the agricultural industry emerging as an option due to the variety of phytochemical compounds produced in their metabolism. In this study, both species presented polyphenolic compounds associated with antifungal and antioxidant activity, including hydroxycinnamic acids, stilbenes, flavonoids, catechins, polyphenols, and methoxycinnamic acids. The results obtained in this work suggest that the plant extracts exhibit inhibitory capacity on *F. oxysporum* f. sp. *lycopersici* development, with an inhibition percentage exceeding 80 % and low IC50 values. This effectiveness is attributed to various phytochemical compounds in both extracts, positioning them as alternatives for the control of *F. oxysporum* f. sp. *lycopersici*. Notably, *V. album* has not been previously reported for its use in directly controlling phytopathogenic microorganisms, making this research the first report of *V. album* ethanolic extract for such purpose.

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