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Shinus molle essential oil against *Aspergillus parasiticus* and *Cladosporium cladosporiodes*

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Abstract

Aspergillus parasiticus and *Cladosporium cladosporioides* are fungi that present significant health risks. This study explored the antifungal potential of *Shinus molle* essential oil, a plant with a rich cultural history of medicinal and environmental uses. The essential oil was extracted from the leaves of *Shinus molle* using hydrodistillation and characterized by gas chromatography. The mean inhibitory concentration (IC_{50}) and minimum inhibitory concentration (MIC) were established based on the radial growth of *A. parasiticus* and *C. cladosporioides*. The IC_{50} of the essential oil was specifically evaluated to assess spore germination. The primary compounds identified in the essential oil of S. molle included a-pinene, a-phellandrene, and γ-elemene. The findings revealed that the essential oil *i*nhibited over 50% of the radial growth of both fungi after 96 hours. However, no significant differences were observed in the IC_{50} values for spore germination at 24 hours compared to the controls. The MIC recorded was notably high for the fungi evaluated. This study suggests that while *S. molle* essential oil does not inhibit the germination of *A. parasiticus* and *C. cladosporioides,* it restricts their growth radial at elevated concentrations.

Keywords: Antifungal, essential oil, fungi

Introduction

Aspergillus parasiticus contaminates crops for human consumption, presenting potential health risks due to its production of toxic and carcinogenic mycotoxins (Valencia-Quintana et al., 2020). Conversely, *Cladosporium cladosporioides* is a saprophytic fungus that thrives in indoor environments and can provoke allergies. It may act opportunistically in certain situations, resulting in serious fungal infections that endanger human health (Alam et al., 2022).

Research has highlighted essential oils' health benefits and antifungal properties (Cai et al., 2021). This study focused on *Shinus molle,* a plant known for its rich cultural history in traditional medicine and its applications in addressing various diseases and environmental concerns (Razzak et al., 2023). The research aimed to evaluate the antifungal potential of *S. molle* essential oil on the radial growth and spore germination of both *A. parasiticus* and *C. cladosporioides*.

Materials and methods

The essential oil was obtained through hydro-distillation of *S. molle* leaves harvested from the Yaqui Valley in Sonora (coordinates: 27°34'39"N, 109°56'19"W). 100 grams of leaves were dried for one week and then finely ground into a powder. The powdered leaves were hydro-distillated using Clevenger equipment (IMPARLAB, Mexico) for 4 hours. Subsequently, the hydrophobic extract was separated via decantation.

The characterization of the essential oil was performed according to Quintana-Obregón et al. (2017) using gas chromatography with a GC-7890B system coupled to 240 selective mass detector (MS) equipped with a 70-eV electrical ionization system (Agilent Technology). The capillary column was a DB-5 (50 m x 0.25 mm) conditioned at 60°C for 10 minutes. The temperature was then gradually elevated at a rate of 20°C per min to 180°C, supported for 2 minutes, and later increased at 4°C per min[ute] to 250°C, where it was held

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for 4.5 minutes. Helium served as the carrier gas, flowing at 2 mL per min. The ionization chamber and transfer line temperatures were set to 220°C and 280°C, respectively. The constituents were identified by comparing their retention indices, calculated through linear interpolation based on the retention times of n-alkanes from C8 to C22, along with mass spectra matched against the NIST 98 database and relevant literature from the National Institute of Standards and Technology.

Strains of A. *parasiticus* (ATCC 16992) and *C. cladosporioides* (isolated from safflower seeds) were incubated on potato dextrose agar (PDA) for 7 days at 25°C under a 12-hour light-dark photoperiod. Following incubation, fungal spores were suspended in sterile Tween 20® (0.1% v/v) and mixed with a magnetic stirrer for five minutes. The concentration of spores per milliliter was quantified using a Neubauer chamber.

The mean inhibitory concentration (IC_{50}) and minimum inhibitory concentration (MIC) were assessed based on the fungi's radial growth. PDA supplemented with 1% Tween 80 was autoclaved and cooled to 45°C, and then the essential oil was added in volumes of 12-15 mL into each Petri dish. The essential oil dilutions tested included 10, 100, 250, 500, and 1000 ppm, while control dishes were prepared with only PDA and PDA with Tween 80 (0 ppm), excluding the essential oil. Next, the center of each Petri dish was drilled using a sterilized Pasteur pipette, and 10^4 spores were introduced into the hole. The dishes were then incubated at 25°C with a 12-hour light-dark cycle, and the radial growth of the mycelium was measured every 12 hours. The concentration of essential oil that inhibited 50% or more of the radial growth (IC_{50}) and the minimum inhibitory concentrations (MIC) were determined using Probit analysis with NCSS 2000 Statistical Software (Number Cruncher Statistical Systems, Utah, USA).

For the IC_{50} assessments of each fungus, Petri dishes were prepared as described earlier, but without creating the hole for spore addition, following the method outlined by López-Meneses et al. (2016).

Three replicates were conducted for radial growth and spore germination, and standard deviations

Table 1. Constituents of the essential oil *S. molle*.

Compound number	Compound name	RI	Relative Proportion(%)
1	α-Thujene	923	1.177
2	α-pinene	**929**	**19.433**
3	Camphene	944	0.15
4	1-Butadine, 2,3,3-trimethyl	969	0.17
5	Sabinene	974	0.471
6	β-pinene	976	1.667
7	α-Phellandrene	**1003**	**17.902**
8	3-carene	1010	0.216
9	Cyclohexene, 1,5,5-trimethyl-3-methylene-	1018	1.613
10	Limonene	1027	6.64
11	β-Phellandrene	1032	6.42
12	γ-Terpinene	1063	1.277
13	Terpinolene	1082	1.689
14	2-Oxabicyclo [2.2.2} octan-6-ol, 1,3,3-trimethyl-, acetate	1322	0.122
15	Elixene	1345	0.509
16	α-Copaene	1387	0.386
17	β-Elemene	1395	8.306
18	β - Caryophyllene	1436	3.237
19	γ-elemene	1444	**12.404**
20	α-Humulene	1460	0.204
21	γ-Muurolene	1472	0.402
22	Germacrene D	1480	0.141
23	α-Selinene	1491	0.352
24	Bicyclogermacrene	1500	7.697
25	α-Bulnesene	1507	0.576
26	γ-cadinene	1515	1.927
27	Caryophyllene oxide	1580	1.231
28	Guaiol	1630	0.473
29	β-Eudesmol	1673	1.274
30	(-)-Spathulenol	1828	0.65

were calculated. The data were analyzed using a one-way factorial design and an analysis of variance (ANOVA) (P≤0.05). Means (n=3) were compared using the Tukey-Kramer test, with a confidence interval of 95%. Statistical analyses were conducted using JMP statistical software version 5.0.1 (SAS, 2001).

Results and Discussion

The chemical composition presumptive of essential oil derived from *S. molle* leaves is shown in **Table 1**. Most compounds were a-pinene, a-phellandrene, and γ-elemene. Also, other terpenoids and compounds are in minor proportion.

The mycelium showed a statistical difference in growth radial at 96 h between essential oil concentrations (**Table 2**) for both fungi. The IC_{50} obtained were 341 and 551 ppm for *A. parasiticus* and *C. cladosporioides,* respectively. MIC was 108, 686, 971 in *A. parasiticus* and 40, 543 ppm for *C. cladosporioides.* No statistical difference (P=≤0.05) was observed in the IC_{50} for spore germination compared to the controls (PDA and PDA-Tween 80)

Conclusion

The findings show that the essential oil of *S. molle* does not impede the germination of the fungal spores of *A. parasiticus* and *C. cladosporioides.* In the mycelial development stage of the fungus, growth is inhibited at doses of 500 ppm; López-Meneses previously observed that high doses of 10,000 ppm are required to inhibit *A. parasiticus*. The inhibition of mycelial growth is owing to alterations in hydrophobicity and interactions with the cell membrane caused by essential oil in the culture media (Wu et al., 2024), although the concentration is elevated. Consequently, the essential oil of *S. molle* is not recommended for managing the fungi *A. parasiticus* and *C. Cladosporiodes.*

Table 2. Growth radial of fungi in PDA medium with essential oil of S. molle at 25°C at 96 hours.

Concentration (ppm)	Radial growth (mm)	
	A. parasiticus	*C. cladosporiodes*
0	24.13±1.24 A	15.67±1.61 A
10	19.50±0.71 AB	11.58±1.13 AB
100	21.83±1.38 A	10.92±1.04 ABC
250	14.00±0.35 CD	9.33±2.96 BC
500	11.83±1.26 D	6.42±2.16 C

Average of three replicates and standard deviation. Capital letters show statistical groups in the column (P≤0.05).

Conflict of interest

None of the authors has a conflict of interest to disclose.

References

Alam, S., Nisar, M., Bano, S.A., Ahmad, T. 2022. Impact of aerial fungal spores on human health. In Ahmed T. & Hashmi M.Z. (ed.) *Hazardous environmental micro-pollutants, health impacts and allied treatment technologies, emerging contaminants and associated treatment technologies*. Springer International Publishing, Cham, Switzerland. p. 219–240.

Cai, J., Yan, R., Shi, J., Chen, J., Long, M., Wu, W., Kuca, K. 2021. Antifungal and mycotoxin detoxification ability of essential oils: A review. *Phytotherapy Research* 36: 1-71.

López-Meneses, A.K., Plascencia-Jatomea, M., Lizardi-Mendoza, J., Rosas-Burgos, E.C., Luque-Alcaraz, A.G., Córtez-Rocha, M.O. 2015. Antifungal and antimycotoxigenic activity of essential oils from *Eucalyptus globulus*, *Thymus capitatus* and *Schinus molle*. *Food Science Technology* 35: 664-671.

Quintana-Obregón, E.A., Sánchez-Mariñez, R.I., Cortez-Rocha, M.O., González-Aguilar, G.A., 2017. In vitro antifungal activity of a mix orange terpenes against *Alternaria tenuissima*. *Revista Mexicana de Micología* 45: 7-12.

Martins, M.R., Arantes, S., Candeias, F., Tinoco, M.T., Cruz-Morais, J. 2014. Antioxidant, antimicrobial and toxicological properties of essential oils. *Journal of Ethnopharmacology* 151: 485-492.

Razzak, A., Khiari, R., Moussaoui, Y., Belgacem, N. 2023. *Schinus Molle*: Currently Status and Opportunity. In: Khiari, R., Jawaid, M., Belgacem, M.N. (ed.) *Annual Plant: sources of Fibres, Nanocellulose and cellulosic derivatives: Processing, Properties and Applications*. Springer Nature, Singapore. p. 185–213

Valencia-Quintana, R., Milić, M., Jakšić, D., Klarić, M.Š., Tenorio-Arvide, M., Pérez-Flores, G.A., Sánchez-Alarcón, J. 2020.Environment changes, aflatoxins, and health issues, a review. *International Journal of Environmental Research and Public Health* 17:7850.

Wu, H., Zhao, F., Li, Q., Huang, J., Ju, J. 2024. Antifungal mechanism of essential oil against foodborne fungi and its application in the preservation of baked food. *Critical Reviews in Food Science and Nutrition* 64: 2695-2707.

Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.