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Impact of Essential Oils Composition and Exposure Methods on Fungal Growth and Morphology: Insights for Postharvest Management

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ABSTRACT

Postharvest fungal diseases caused by *Colletotrichum*, *Rhizopus*, and *Penicillium* are major concerns, driving the search for sustainable antimicrobial alternatives to enhance food shelf life. This study examines the chemical composition of essential oils (EO) from *Origanum vulgare*, *Syzygium aromaticum*, *Cymbopogon citratus*, *Cymbopogon martinii*, *Mentha piperita*, and *Mentha spicata*, evaluating their in vitro effectiveness against *Colletotrichum* sp., *Rhizopus stolonifer, and Penicillium expansum*. Different EO concentrations were tested via volatile exposure and direct contact to determine the minimum inhibitory concentration (MIC) for each fungus. The results indicate that there is no universal strategy for prevention and control, as the effectiveness of EO depends directly on the fungal species. *Colletotrichum* sp. and *R. stolonifer* were more susceptible to volatiles from *O. vulgare* (200 µL/L_air) and *M. piperita* (180 µL/L_air), respectively, whereas *P. expansum* was more sensitive to direct contact with *O. vulgare* (250 µL/L_medium). Scanning electron microscopy (SEM) revealed that *O. vulgare*, rich in phenolic terpenes, and *C. citratus*, rich in aldehydic monoterpenes, induced hyphal breakage and twisting at varying intensities in these three common postharvest fungi. The results highlight the potential of EO via volatile exposure and direct contact as a promising alternative for postharvest fungal control.

1 | Introduction

The consistent supply of high-quality fresh and safe fruit to domestic and international consumers is a significant challenge in postharvest technology. Food losses occur due to several reasons, such as inadequate storage conditions, refrigeration, pest attacks, microbial contamination, fruit and vegetable diseases, packaging, distribution infrastructure, and other factors (Dora et al. 2021; FAO 2015). Postharvest losses can be severe, reaching up to 60% of the production of certain products in extreme cases. Among the causes, biological damage caused by pests and diseases is estimated to cause more than 40% of total

losses in fruits and vegetables. Particular emphasis should be given to disease-causing fungi such as *Colletotrichum*, *Rhizopus,* and *Penicillium* (Karoney et al. 2024).

Colletotrichum sp. infects various fruit crops including apples, strawberries, and mangoes (Galsurker et al. 2018). Its presence can lead to rotting and decay of the fruit, resulting in significant losses during postharvest handling and storage (Zhao et al. 2022). *Rhizopus stolonifer* commonly infects soft fruits such as strawberries and it is the cause of soft rot (Bertolo et al. 2025). This fungal infection can spread rapidly and cause extensive damage to the harvested fruit, leading to significant losses in quantity and quality since the losses can reach 50% of the production (Ventura-Aguilar et al. 2021; Zhao et al. 2022). Another common fungus in postharvest is *Penicillium expansum,* which infects apples and pears, causing a disease known as blue mold (Nybom et al. 2020). This fungal infection can result in the deterioration of the fruit's appearance, texture, and taste, making it unsuitable for consumption or sale (Jijakli and Lepoivre 2004; Saleh and Al-Thani 2019; Zhao et al. 2022). Traditionally, these diseases are controlled with synthetic chemical fungicides. However, this control method carries the risk of residual contamination in the environment and of perishable products, which can harm human and animal health (Carmona-Hernandez et al. 2019).

Alternative plant-based control methods with fungicidal and fungistatic properties are gaining importance due to their advantages such as green safety, health, and environmental protection. In this context, essential oils (EO) stand out for their potential action against phytopathogenic fungi (Achimón et al. 2021; Duan et al. 2024; Lin et al. 2023; Maurya et al. 2021; Reyes-Jurado et al. 2020; Zang et al. 2023). In aromatic plants, structures like seeds, buds, flowers, fruits, leaves, twigs, bark, and roots produce these compounds through secondary metabolism as a defense mechanism against predators and diseases. Furthermore, these metabolites have also demonstrated medicinal, antioxidant, insecticidal, and antibacterial properties. In addition, EO such as oregano, clove, lemongrass, spearmint, and peppermint are classified as generally recognized as safe (GRAS) and exhibit greater sensory acceptance (Al-Maqtari et al. 2022; Almeida et al. 2024).

EO compositions are a mixture of many chemical compounds, with characteristics predominantly hydrophobic and with low molecular weight (Al-Maqtari et al. 2022; Duan et al. 2024; Zang et al. 2023). The presence of aliphatic alcohols, phenolics, terpenes, acids, and flavonoids in major concentrations contributes to strong antimicrobial effects (Al-Maqtari et al. 2022; Zang et al. 2023). For antifungal applications, EO molecules in the aqueous phase can form micelles, restricting availability and reducing action potential for aerial mycelium, but in the vapor phase, molecules are free to act (Reyes-Jurado et al. 2020).

The functional groups of the major and minor compounds found in EO play a crucial role in their antimicrobial activity. Generally, microorganisms are affected by the constituents of plant EO in two distinct ways. First, molecules can alter the composition of microbial cells and mycelium, leading to changes in organelles, cell membranes, and cell walls, resulting in physical changes in morphological structures. Second, these oils can reduce or suppress spore production and germination (Abdi-Moghadam et al. 2023).

The ability of EO components to permeate the cell wall and disrupt the cell membrane structure through interactions with lipids, proteins, or porins can lead to cytoplasmic leakage, disruption of wall synthesis, interference with enzymatic reactions, ATPase pump damage, and cytoplasmic acidification. These mechanisms ultimately result in cell lysis and death (Abdi-Moghadam et al. 2023; Duan et al. 2024; Lin et al. 2023). These modes of action highlight the potential applications of EO in postharvest settings, as they can effectively inhibit the development of pathogenic fungi in fruits and vegetables, ensuring greater stability and quality.

Aligned with Goal 2.4, 'sustainable food production and resilient agricultural practices' proposed by the United Nations General Assembly, as well as the FAO Save Food: Global Initiative on Food Loss and Waste Reduction (FAO 2015), this study aimed to evaluate the antifungal efficacy of EO across different application methodologies, with a focus on their potential real-world application in postharvest technologies. Two primary application methods were investigated: (1) direct contact, which could support the incorporation of EO into edible coatings, and (2) volatile exposure, which could guide the development of active packaging solutions. Due to their safety, sensory compatibility, commercial accessibility, and previous data in the literature, oregano, clove, lemongrass, palmarosa, peppermint, and spearmint EO were selected to evaluate and compare the antifungal activity by direct contact and exposure to volatiles against *Colletotrichum* sp., *Rhizopus stolonifer,* and *Penicillium expansum*.

2 | Material and Methods

2.1 | Materials

EO of *Origanum vulgare* L. (oregano), *Syzygium aromaticum* L. (clove), *Cymbopogon martinii (Roxb.) J. F. Watson* (palmarosa), and *Mentha piperita* L. (peppermint) were obtained from Harmonie Aromaterapia (Florianopolis, SC, Brazil); *Mentha spicata* L. (spearmint) EO was acquired from Terraflor Aromaterapia (Alto Paraiso de Goias, GO, Brazil); and *Cymbopogon citratus (DC.) Stapf* (lemongrass) EO was acquired from Mundo dos Óleos (Brasilia, DF, Brazil). The method of extraction of the essential oils was steam distillation (leaves).

The fungal strains used in antifungal activity assays were *Colletotrichum* sp. CBMAI 0864 *provided by* Coleção Brasileira de Micro-organismos de Ambiente e Indústria – CBMAI/UNICAMP, *Rhizopus stolonifer* CCT 0276 NRRL 1478 obtained from Fundação Tropical de Pesquisas e Tecnologia André Tosello, and *Penicillium expansum* CMIIAA PEN 001 provided by Embrapa Agroindústria de Alimentos.

2.2 | Essential Oil Chemical Composition Analysis

The essential oils were characterized by gas chromatography–mass spectrometry (GC–MS) as Fukuyama et al. (2024)

described. A Shimadzu GC–MS 2010 Plus system (Kyoto, Japan) equipped with a quadrupole mass spectrometer (GC–MS) was used. The EO components were separated using a DB-5MS capillary column (30 $m \times 0.25$ μm $\times$ 0.25 mm) with helium as the carrier gas at a 1 mL/min flow rate. Amber vials containing 10% (v/v) essential oil in dichloromethane were prepared immediately before injection. The samples were then injected (1 μL) into the GC–MS using the Shimadzu AOC20i autosampler system. The chromatographic conditions were as follows: injector temperature: 250° C (split mode 1:50); oven temperature program: starting at 60° C (1 min), followed by an increase of 3° C/min up to 240° C; interface and ion source temperature, 240° C; EI +70 eV; and mass range, m/z 35–350. The linear temperature programmed retention index (RI) was calculated using an alkane solution (C8–C20). The identification of the analytes was performed as described by Spencer et al. (2021), comparing the RI and mass spectra obtained for the sample with mass spectra and RI of the literature, with the similarity of at least 85% for the mass spectra, and maximum variation in RI of ±10. The identification of analytes was confirmed by the co-injection of authentic standards whenever these were available. The semiquantitative analysis of essential oils (% relative area) was performed in the same gas chromatography system but using the flame ionization detector (GC-FID). All qualitative and semiquantitative analyses were performed in triplicate.

2.3 | Antifungal Activity of Essential Oils by the Volatile Exposure Method

Antifungal assays from volatile EO were adapted from Aguilar-González et al. (2015). Potato dextrose agar (PDA) medium from Kasvi was prepared and plated in a petri dish in a manner that contained 20 mL of medium and 50 mL of air. Filter paper discs (2 cm diameter) were placed in the center of the petri dish lid (Figure S1A) and different volumes of EO, ranging from 1 to 100 μL (equivalent to 20 μL/L_{air} to 2000 μL/L_{air}) were applied to the paper disc (Figure S1B). A control group without essential oil added to the filter paper was established. A concentration limit of 2000 μL/L air was stabilized due to the retention capacity of the filter paper in the lid. All treatments were performed in quadruplicate. A 9-mm diameter fungal agar plug was placed in the center of each PDA petri dish as the inoculum. The plates were incubated at 28 ºC for 7 days, and fungal growth was monitored every 24 h.

2.4 | Antifungal Activity of EO by the Direct Contact Method

Potato dextrose agar (PDA) medium from Kasvi with Tween 80 (0.5% v/v) was prepared containing different EO concentrations: 62.5, 125, 250, 500, 750, and 1000 μL/L, and the treatment without the addition of EO was used as a control. A threshold concentration of 1000 μL/L was set due to the solubility of the EO in the culture medium with the surfactant (higher concentrations resulted in separation and accumulation of the oil on the surface of the medium). The inoculation was done by transferring agar plugs (9 mm diameter) from an actively growing colony. All treatments were carried out in triplicate. The incubation temperature was 28 ºC for 7 days, and the fungal growth was monitored every 24 h.

2.5 | Scanning Electron Microscopy (SEM)

2.5.1 | Fungi Exposed to EO Volatiles

Sample preparation followed the method of Yu et al. (2022) with some modifications. Potato dextrose agar (PDA) medium from Kasvi was inoculated with a 9-mm diameter agar plug containing fungal mycelium (*Colletotrichum* sp., *Rhizopus stolonifer*, or *Penicillium expansum*) and then incubated at 28 ºC for 4 days. For the volatile exposure method, EO of *Origanum vulgare* and C. *citratus* were individually applied to a paper filter disc placed on the petri dish lid. These EO were selected because they contain major components with distinct chemical groups, specifically alcohol and aldehyde, respectively. The EOs were used at concentrations corresponding to the minimum inhibitory concentration (MIC) and at higher concentrations. The tested volatile concentrations for *Colletotrichum* sp. were 200 and 2000 μL/L_{air} for *O. vulgare* EO and 1200 and 2000 μL/L_{air} for *Cymbopogon citratus* EO. For *Rhizopus stolonifer*, the concentrations were 200 and 400 μL/μL/L_{air} for *O. vulgare* EO and 400 and 600 μL/L_{air} for *C. citratus* EO. For *Penicillium expansum*, 400 and 800 μL/L_{air} were tested for both EO.

2.5.2 | Fungi Exposed to EO Through Direct Contact

Using the same incubation period previously described, *O. vulgare* and *C. citratus* EO were added to the medium at concentrations corresponding to the minimum inhibitory concentration (MIC) obtained for each fungus and at concentrations exceeding the MIC. For *Colletotrichum* sp., *Oregano vulgare* EO was tested at 750 and 1000 μL/L, and *Cymbopogon citratus* EO was tested at 1500 and 2000 μL/L. For *Rhizopus stolonifer*, *Oregano vulgare* OE was tested at 250 and 1000 μL/L, and *Cymbopogon citratus* EO was tested at 750 and 1000 μL/L. For *Penicillium expansum*, *Oregano vulgare* EO was tested at 250 and 1000 μL/L, and *Cymbopogon citratus* EO was tested at 1000 and 1500 μL/L.

2.5.3 | Stubs Preparation

Both tests (volatile exposure and direct contact methods) were carried out in triplicate considering the control group (without adding the EO). After 48 h of incubation, a disc (5 mm in diameter) was removed and placed in glutaraldehyde (3% v/v) overnight and then immersed in a phosphate buffer (0.05 M, pH 6.8). Samples were dehydrated in a graded series of acetone concentrations (30%, 50%, 70%, 90%, and 100% v/v) and dried in liquid carbon dioxide at the critical point. The dried discs were deposited on stubs with adhesive tape and coated with gold for observation in the scanning electron microscope (SEM - SEM JEOL JSM-6701F, Tokyo, Japan).

3 | Results and Discussion

3.1 | EO Composition

The composition of the EO used in this study is presented in Table 1. The corresponding chromatograms are available in the Supplementary Materials, listed as follows: Figure S2—*C. citratus* (Lemongrass) EO; Figure S3—*E. caryophyllus* (Clove) EO; Figure S4—*O. vulgare* (Oregano) EO; Figure S5—*M. piperita* (Peppermint) EO; Figure S6—*M. spicata* (Spearmint) EO; and Figure S7—*Cymbopogon martini* (Palmarosa) EO.

Origanum vulgare L. EO presented carvacrol (79.93%), thymol (5.00%), *p*-cymene (4.81%), and γ-terpinene (4.28%) as the main components, respectively. These results resemble the data of Duan et al. (2024), which presented those compounds as the main components (carvacrol 66.01%; thymol 3.51%; *o*-cymene 3.03%; and γ-terpinene 2.54%). However, the concentrations of EO components can vary due to several factors, intrinsic and extrinsic to the plant of origin. As an example, for *Origanum vulgare* ssp. *hirtum*, the levels of carvacrol and γ-terpinene are higher in the upper leaves (Chizzola 2013). Carvacrol and thymol are phenolic compounds with antifungal activity (Leonelli Pires de Campos et al. 2022). In *Aspergillus flavus,* there is evidence that exposure to carvacrol can cause damage to the plasma membrane, DNA damage, a decrease in mitochondrial membrane potential, and oxidative stress due to reactive oxygen species accumulation (Duan et al. 2024).

Syzygium aromaticum EO had phenolic compounds eugenol and caryophyllene as the main components, representing, respectively, 84.49% and 11.68% of the EO composition. The main components observed agree with other studies, such as Achimón et al. (2021) with *S. aromaticum* EO presenting 88.70% of eugenol and 6.55% caryophyllene. This research also reports that the polarity of eugenol favors the ability to penetrate cell walls, interacting and causing greater damage to the membrane. The inhibitory activity of this compound has already been observed against animal and plant pathogens, food poisoning, spoilage bacteria, and fungi such as *Fusarium verticillioides and Rhizopus stolonifer* (Achimón et al. 2021; Chizzola 2013; Zhou et al., 2017).

C. martinii EO presented geraniol (86.39%) as the main component as shown by Devi et al. (2021) and Sen et al. (2023). Geraniol occurs mainly in *Cymbopogon* species (as citronella *C. winterianus* and *C. nardus)*, presenting minty, floral, and lemon-like odor (Chizzola 2013; Devi et al. 2021). The antifungal properties of geraniol are related to the interaction with the membrane and activation of apoptosis-related genes. Among the responses already reported to exposure, ATP-dependent efflux is associated with a loss of the mitochondrial membrane potential, an increase in membrane fluidity, cell wall rupture, reduced hyphal development, and increased oxidative activity (Lin et al. 2023; Scariot et al. 2021). Furthermore, this EO has antifungal properties already reported for several fungi, such as *Trichophyton rubrum*, *Aspergillus fumigatus*, *Alternaria solani*, *Aspergillus niger, Aspergillus flavus*, and *Fusarium oxysporum* (Devi et al. 2021; Lin et al. 2023; Sen et al.; 2023).

C. citratus EO presented neral/α-citral (44.59%), geranial/β-citral (35.99%), and geraniol (8.42%) as the main components. These components were also observed as the main components of *C. citratus* EO in other studies, such as in Plata-Rueda et al. (2020) and Madi et al. (2021). This EO has an antifungal activity that can be associated with its major compounds neral, geranial, and geraniol, showing the effect on some fungi, such as *Aspergillus niger, Botrytis cinerea, Cladosporium herbarum, Colletotrichum coccodes,* and *Rhizopus stolonifer* (Lee et al. 2020). Fungi *Raffaelea quercus-mongolicae* and *Rhizoctonia solani* treated with *C. citratus* produce intracellular reactive oxygen species (ROS) that, when accumulated, can induce cellular apoptosis. Besides that, neral and geranial can cause cell membrane damage and are associated with a negative regulation of the biosynthesis of ergosterol, a component of the cell membrane in *Penicillium digitatum* (Lee et al. 2020).

M. piperita EO presented menthol (36.59%) and menthone (36.59%) as the main components. Giménez-Santamarina et al. (2022) and Hudz et al. (2023) also describe these compounds as the majority in their research. However, Samber et al. (2015) found menthol (34.82%), carvone (19.54%), and menthone (9.10%) as the main components. The possible antifungal mechanisms of action of this EO are related to the decrease in PM-ATPase activity, the ergosterol content, and cell membrane breakage (Hudz et al. 2023; Samber et al., 2015). Moreover, the antifungal effects of *Mentha peperita* oil have already been reported for *Candida albicans*, *Candida tropicalis*, *Candida glabata*, *Fusarium moniliforme*, *Aspergillus niger*, and *Aspergillus fumigates (*Giménez-Santamarina et al. 2022; Hudz et al. 2023; Samber et al., 2015).

M. spicata EO showed carvone (66.54%) and limonene (25.34%) as the main components as also demonstrated by Giménez-Santamarina et al. (2022) and Mansoori et al. (2022). Carvone's antifungal effect is closely associated with the change in the cellular rigidity of the hyphae due to the molecule's high penetration capacity. Furthermore, as already demonstrated in *Fusarium*, inside the cell, it interacts with the enzyme system and acts to destabilize mitochondrial functions. Limonene also acts to increase permeability and damage the cell membrane, with consequent ionic dysregulation. In *C. tropicalis*, the extravasation of cytoplasmic proteins and changes in ATP synthesis pathways are also reported (Bouyahya et al. 2021; Soliman et al. 2022). The antifungal activity of *M. spicata* EO was described for *Fusarium* spp., *Aspergillus* spp., *Penicillium* spp, and *Candida tropicalis* (Bouyahya et al. 2021; Soliman et al. 2022; Yu et al. 2022).

3.2 | Antifungal Assays

3.2.1 | Antifungal Activity of EO Volatile Compounds

Figure 1 presents the minimum inhibitory concentration (MIC) data observed for each volatile EO against the fungi *Colletotrichum* sp., *Rhizopus stolonifer*, and *Penicillium expansum*.

On *Colletotrichum* sp. growth control, O. *vulgare* presented the lowest MIC (200 $\mu L/L_{air}$), followed by *S. aromaticum* volatile EO (400 $\mu L/L_{air}$). *C. martinii* and *M. spicata* presented high MIC values; meanwhile, none of the volatile EO concentrations analyzed for *C. citratus* and *M. piperita* were able to inhibit

TABLE 1 | Identification of EO volatile compounds by GC–MS and GC-FID.

Compound	RI exp	RI lit	% Relative area					
			OV	*SA*	*CM*	*CC*	*MP*	*MS*
α-Thujene	925	929	0.64	—	—	—	—	—
α-Pinene	932	934	0.35	—	—	—	0.80	0.43
Camphene	948	952	—	—	—	0.38	—	—
Sabinene	974	974	—	—	—	—	—	0.30
β-Pinene	978	977	—	—	—	—	0.82	—
Sulcatone	984	986	—	—	—	0.49	—	—
β-Myrcene	989	992	0.91	—	—	—	—	0.57
α-Terpinene	1016	1017	0.57	—	—	—	—	—
p-Cymene	1023	1024	4.81	—	—	—	—	—
Limonene	1028	1027	—	—	—	0.36	—	—
Eucalyptol	1034	1034	—	—	—	—	7.23	—
Limonene	1035	1035	—	—	—	—	—	25.34
trans-β-ocimene	1047	1047	—	—	0.91	—	—	—
γ-Terpinene	1057	1059	4.28	—	—	—	—	—
Linalool	1100	1100	—	—	1.41	0.36	—	—
Citronella	1152	1150	—	—	—	0.63	—	—
Menthone	1165	1163	—	—	—	—	36.69	—
Isomenthol	1173	1177	—	—	—	—	8.31	—
Terpinen-4-ol	1179	1179	—	—	—	—	—	0.49
Menthol	1189	1181	—	—	—	—	36.59	—
trans-Dihydrocarvone	1199	1199	—	—	—	—	—	1.08
Nerol (*cis*-geraniol)	1230	1228	—	—	—	1.08	—	—
Pulegone	1244	1244	—	—	—	—	0.81	—
Geranial/β-Citral	1245	1242	—	—	—	35.99	—	—
Piperitone	1258	1259	—	—	—	—	0.65	—
Carvone	1261	1254	—	—	—	—	—	66.54
Geraniol	1266	1265	—	—	86.39	8.42	—	—
Neral/α-citral	1275	1277	—	—	—	44.59	—	—
Thymol	1293	1292	5.00	—	—	—	—	—
Isomenthol acetate	1299	1305		—	—	—	3.70	—
Carvacrol	1306	1300	79.93	—	—	—	—	—
Eugenol	1367	1358	—	84.49	—	—	—	—
Geraniol acetate	1386	1385	—	—	7.74	5.34	—	—
β-Bourbonene	1390	1386	—	—	—	—	—	0.73
Caryophyllene	1422	1419	1.67	11.68	1.00	0.45	—	—
α-Caryophyllene	1457	1456		1.58	—	—	—	—
β-Bisabolene	1510	1509	0.34	—	—	—	—	—
Eugenol acetate	1528	1525		0.62	—	—	—	—
Caryophyllene oxide	1586	1583	0.36	0.94		0.36	2.15	0.49
trans-Farnesol	1724	1722	—	—	0.54	—	—	—
Geranyl caproate	1756	1755	—	—	0.61	—	—	—
Total identified			98.86	99.31	98.6	98.45	97.75	95.97

Note: Compounds listed in order of elution from the DB-5MS capillary column; RI exp, retention index determined in the DB-5MS column using a series of C8-C20 alkanes; RI lit, retention index according to the National Institute of Standards and Technology (NIST) and/or Pherobase (https://pherobase.com/).
Abbreviations: CC, *C. citratus*; CM, *C. martinii*; MP, *M. piperita*; *MS*, *M. spicata* (–) *not detected*; OV, *O. vulgare*; SA, *S. aromaticum*.

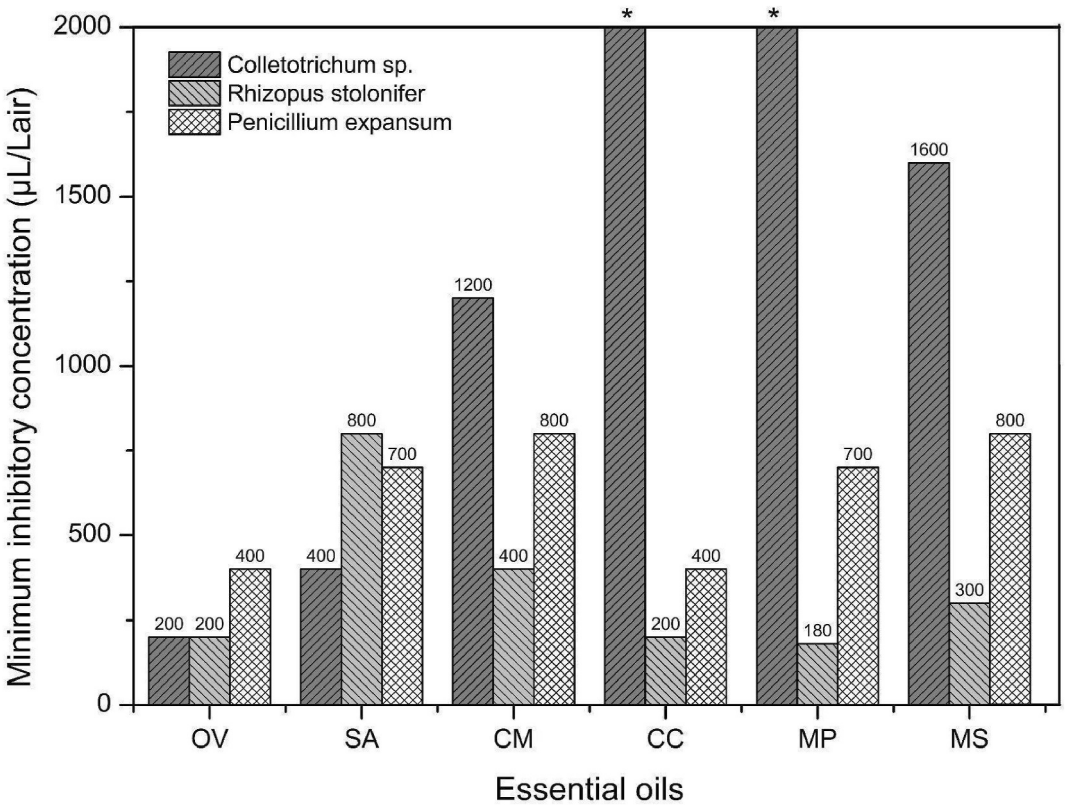

FIGURE 1 | **Minimum inhibitory concentration (MIC) of essential oil Volatiles against phytopathogenic fungi.** CC: *Cymbopogon citratus* EO; CM: *Cymbopogon martinii* EO; MP: *Mentha piperita* EO; MS: *Mentha spicata* EO; OV: *Origanum vulgare* EO; SA: *Syzygium aromaticum* EO. (*) Indicates no antifungal activity observed at the maximum concentration analyzed.

Colletotrichum sp. growth, being that its MIC is superior to 2000 µL/L_{air}. The lowest MIC value obtained for *R. stolonifer* was 180 µL/L_{air} by *M. piperita* EO, followed by *O. vulgare and C. citratus* EO volatiles (200 µL/L_{air}). In opposition, *S. aromaticum* EO presented the highest MIC value to this fungus (800 µL/L_{air}). On the growth control of *P. expansum, O. vulgare* and *C. citratus* had the lowest MIC of 400 µL/L_{air}.

In a pooled analysis of fungal inhibition, *O. vulgare* volatiles were the most efficient in inhibiting the three fungi analyzed, once it presented the lowest MIC for *Colletotrichum* sp. and *Penicillium expansum*, and a great performance for *Rhizopus stolonifer*. *O. vulgare* EO has carvacrol as the main component, which can cause damage to the cell membrane and oxidative stress due to an upregulation of chitinase syntheses, the enzyme that degrades chitin (cell wall component in some fungi, such as *Penicillium* sp.) (Duan et al. 2024; Nunes and Philipps-Wiemann 2018). Similarly, *S. aromaticum* EO, whose major component is also a phenolic alcohol, also presented great inhibitory activity by volatile exposure to the analyzed fungi. The free OH group in the structure of aromatic terpenoids acts as the hydrophilic portion that increases its solubility and is available to form hydrogen bonds they can interact with active sites of different enzymes, thus interrupting their activity (Achimón et al. 2021; Zhou et al., 2017).

R. stolonifer had the lowest MIC value obtained, being a sensitive fungus to the inhibitory activity of volatile EO, especially EO with nonaromatic major compounds, such as *M. piperita, C. citratus,* and *M. spicata*. On the contrary, *Colletotrichum* sp. presented a high MIC value to EO tested, being that terpenes with alcoholic major compounds were more effective in inhibiting fungal growth, especially aromatic monoterpene EO, as *O. vulgare* and *S. aromaticum*. *P. expansum* had an intermediate sensibility to volatile EO and the best results came from phenolic alcohol (*O. vulgare*) and acyclic aldehyde (*C. martinii*) EO.

3.2.2 | Antifungal Activity of EO by Direct Contact Method

Figure 2 shows the EO minimum inhibitory concentration (MIC) using the direct contact method. As shown in exposure to volatiles, *O. vulgare* also had the lowest MIC of all EO tested for the three fungi in direct contact. The MIC values for each fungus were 750 µL/L, 250 µL/L, and 250 µL/L for *Colletotrichum* sp., *Rhizopus stolonifer,* and *Penicillium expansum*, respectively. However, unlike the volatile analysis, *S. aromaticum* had the second-best activity against *R. stolonifer* and *P. expansum* (500 µL/L for both).

The two most efficient EO, *O. vulgare and S. aromaticum,* for the three strains investigated had their composition based on phenolic alcohol. In addition, *Colletotrichum* sp. was the least susceptible to EO investigated, presenting the highest MIC values. This fungus was unable to achieve the MIC value of some EO that had low inhibitory activity, especially EO with a ketone, aldehyde, and cyclic alcohol as the main compounds, such as *C. citratus*, *M. piperita,* and *M. spicata.*

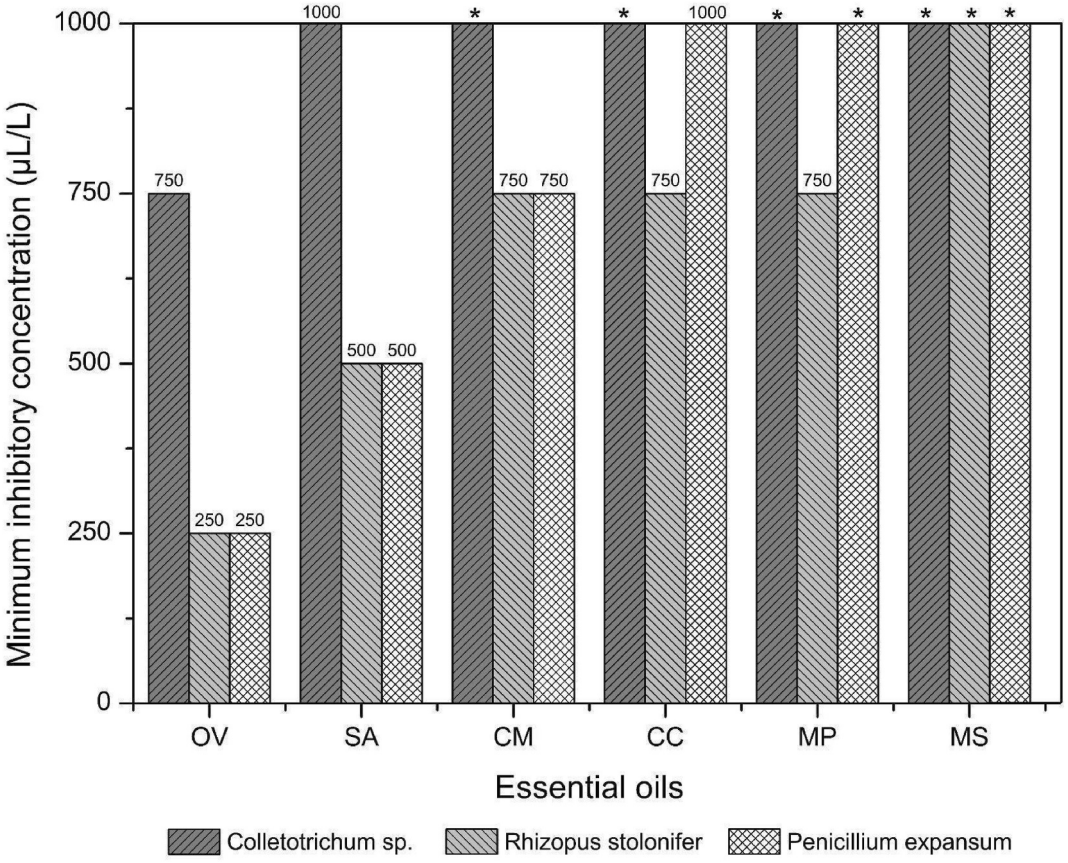

FIGURE 2 | **Minimum inhibitory concentration (MIC) of essential oils in direct contact with phytopathogenic fungi.** CC: *Cymbopogon citratus* EO; CM: *Cymbopogon martinii* EO; MP: *Mentha piperita* EO; MS: *Mentha spicata* EO; OV: *Origanum vulgare* EO; SA: *Syzygium aromaticum* EO. (*) Indicates no antifungal activity observed at the maximum concentration analyzed.

As for the volatile exposure method, *R. stolonifer* was the most sensible for the antifungal activity of EO by the direct contact method. Direct contact highlights the low inhibitory activity for ketone, aldehyde, and cyclic alcohol compounds for this fungus (as observed for *Colletotrichum* sp.).

It was possible to compare the volatile exposure method to the direct contact method by comparing the volume of EO required per petri dish to completely inhibit fungal growth. Thus, it was verified that for *R. stolonifer* and *P. expansum*, the volatile exposure method required less volume of EO with the direct contact method, being that EO with phenolic and acyclic alcoholic major compounds, *O. vulgare*, *S. aromaticum,* and *C. martinii*, respectively, were more efficient in controlling fungal growth. Nevertheless, *Colletotrichum* sp. inhibition required less volume of EO with the volatile exposure method, being that EO with phenolic alcohol was more efficient.

EO demonstrated different results by volatile exposure and direct contact methods due to the different mechanisms of action of each EO and the different structures of each fungus, which influence the susceptibility to the EO (Andrade-Ochoa et al. 2021). In line with the data obtained in this work, other reports describe that EO volatiles may be more effective when compared to direct contact (Álvarez-García et al. 2023; Oliveira Filho et al. 2021; Reyes-Jurado et al. 2020; Tullio et al. 2007). Furthermore, these in vitro results provide the basis for the development of new technologies aimed at mitigating losses due to diseases in fruits and vegetables postharvest. This aligns with the SDG goal 2.4 and The Global Initiative on Food Loss and Waste Reduction (FAO 2015).

3.3 | Scanning Electron Microscopy (SEM)

The morphological effects caused by *O. vulgare* and *C. citratus* EO on fungi are presented in response to the two exposure methods (volatile and direct contact). The images were organized in Figures 3 and 4 for *Colletotrichum* sp., Figures 5 and 6 for *Rhizopus stolonifer,* and Figures 7 and 8 for *Penicillium expansum*. The control without exposure to EO is presented in micrographs (A). Images (B) and (C) demonstrate the effect of the oils when applied at the MIC concentration (based on data in Figures 1 and 2), and the images (D) and (E) present micrographs of fungi exposed to concentrations above the MIC. Furthermore, in Supplementary Materials, each fungus is presented with different magnifications (lines) aiming to highlight the specificities of the structure and size of each species studied.

Common to all controls, the images depict smooth, plump, and neat hyphae. Meanwhile, the presence of EO led to expressive morphological alterations. The initial change observed in

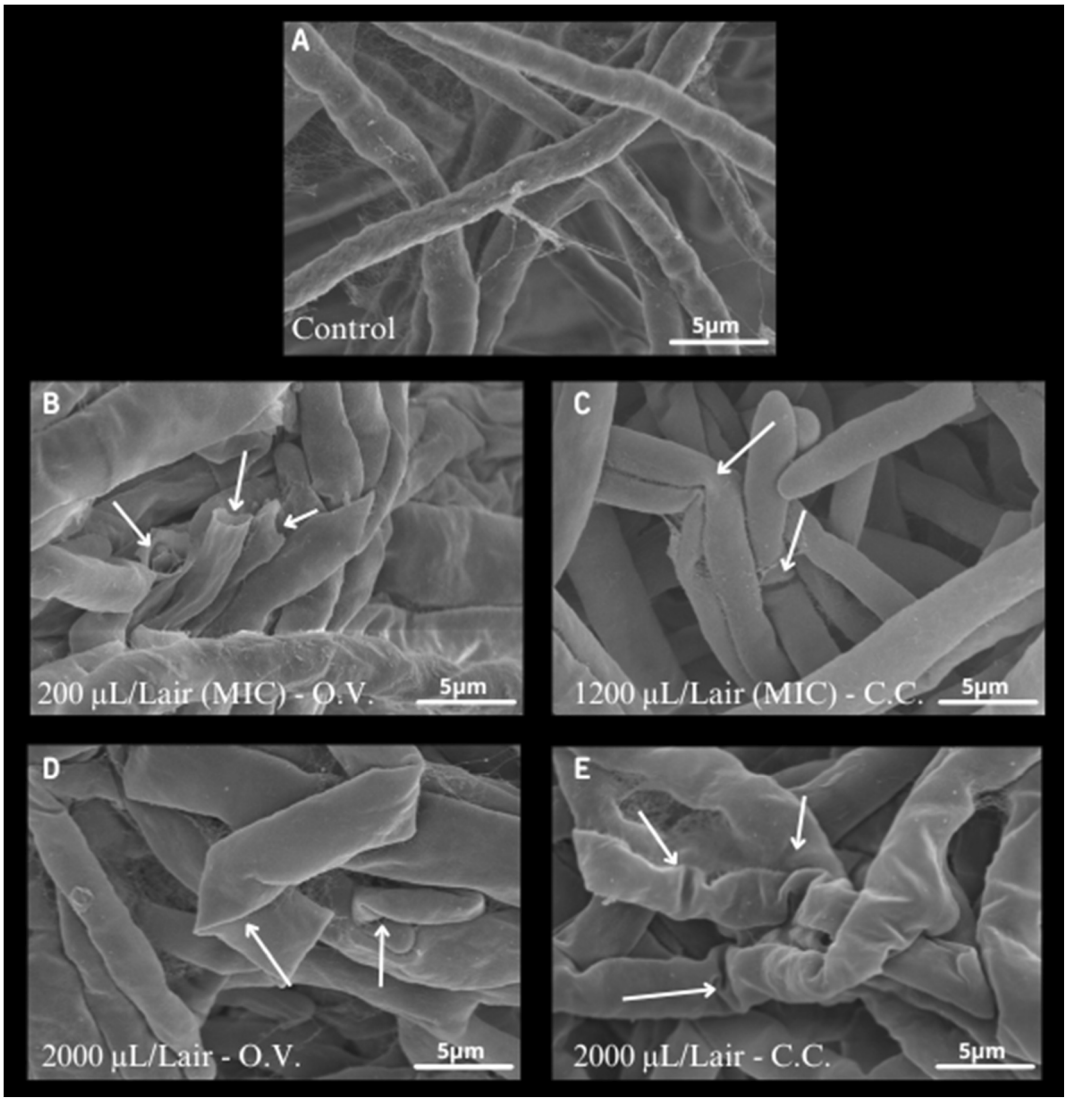

FIGURE 3 | SEM morphological observations of *Colletotrichum* sp. exposed to volatile compounds from *Origanum vulgare* and *Cymbopogon citratus* essential oils. (A) Control—no exposure to essential oils (EOs). (B) Exposed to 200 µL/L $_{air}$ of *Origanum vulgare* EO (**MIC**). (C) Exposed to 1200 µL/L $_{air}$ of *Cymbopogon citratus* EO (**MIC**). (D) Exposed to 2000 µL/L $_{air}$ of *O. vulgare* EO. (E) Exposed to 2000 µL/L $_{air}$ of *C. citratus* EO. All images were captured at 5000× magnification.

Colletotrichum sp. concerning the oils tested is the density of the mycelium (Figures 3 and 4). Control presents spacing between the hyphae (A), whereas the other treatments present a more compact arrangement. This densification of the mycelia is related to the increase in the width of the hyphae (easily comparable between 1B and other micrographs of the same magnitude in Figures S8 and S9 in Supplementary Materials), reducing the free spaces as observed in the control. On the other side, demonstrating the greater resistance of this fungus to *C. citratus* EO, there is no apparent increase in the diameter of the hyphae when exposed to MIC concentration in direct contact (Figure 4), and consequently, low density of the mycelium is observed.

The effect of *O. vulgare* evidenced by the two methods analyzed resulted in the deformation of the *Colletotrichum* sp. hyphae (Figures 3 and 4), making them withered, twisted, and folded. Breakage was observed at the ends of the hyphae with a hollow interior (3 - B) at the lowest MIC concentration (200 µL/L_{air}) for volatiles.

In direct contact, ruptures were more evident at a concentration of 1000 µL/L (4 - D). Although there is no discernible emphasis on the increase in hyphae thickness when exposed to direct contact at a concentration of 1500 µL/L of *C. citratus*, other specific morphological changes are observable, such as the deformation and breakage of the ends (evidenced in 4 - C). For the highest concentration studied, *C. citratus* (2000 µL/L for volatiles and direct contact), the effects were similar to those observed for *O. vulgare*, but with much more evident folds (E in Figures 3 and 4).

Confirming the greater influence of *O. vulgare* on the predictions of *R. stolonifer* (Figures 5 and 6), the mycelium morphology was markedly altered in both methodologies and in both

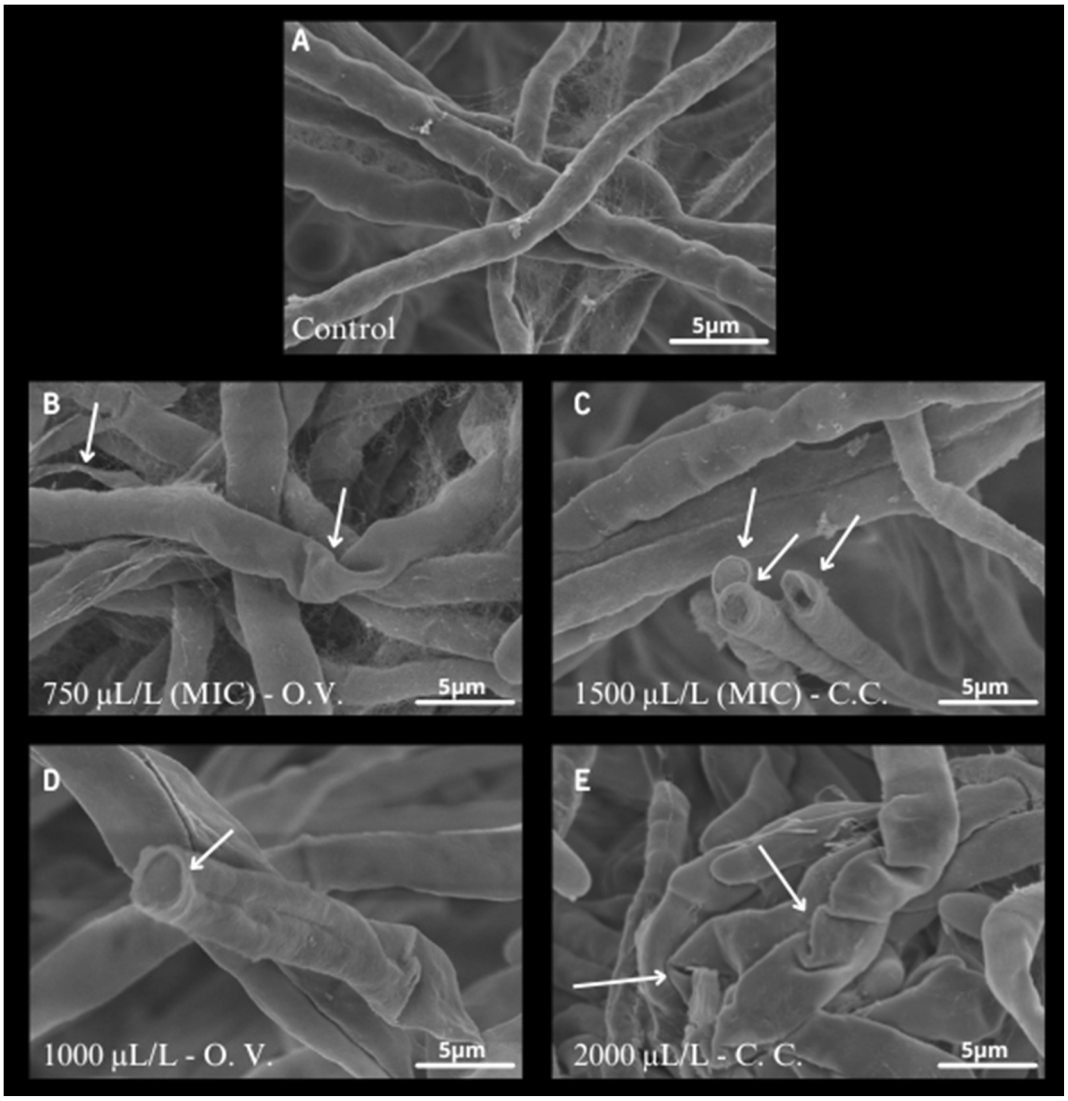

FIGURE 4 | **SEM morphological observation of *Colletotrichum* sp. exposed to direct contact with *Origanum vulgare* and *Cymbopogon citratus* essential oils.** (A) Control—no exposure to essential oils (EOs). (B) Exposed to 750 µL/L of *O. vulgare* EO (**MIC**). (C) Exposed to 1200 µL/L of *C. citratus* EO (**MIC**). (D) Exposed to 2000 µL/L of *O. vulgare* EO. (E) Exposed to 2000 µL/L of *C. citratus* EO. All images were captured at 5000× magnification.

concentrations. At the lowest magnification, the densification of the mycelium is observed when exposed to 1000 µL/L of oil in direct contact (Figure S11—3A compared with S10–3A in the Supplementary Material). However, this characteristic is not associated with the increase in the hyphae diameter as observed in *Colletotrichum* sp. Furthermore, it is possible to observe that under the action of volatiles at the MIC concentration, abrupt breaks with exposure of the interior of the hyphae are observed (Figure 5 - B), as it happens for concentrations above the MIC in direct contact (Figure 6 - D). In addition, the hyphae presented a ribbon shape with folds in the MIC concentration in direct contact (Figure 6 - B) and above the MIC under the action of volatiles (Figure 5 - D).

Although *R. stolonifer* is the fungus most susceptible to the effects of the tested oils, when exposed to *C. citratus,* the morphological changes were subtler. Under the MIC concentration of the volatiles, there were breaks (Figure 5 - C), and above the MIC concentration, the hyphae showed breaks and greater surface roughness (Figure 5 - E). At the MIC value, *R. stolonifer* exposed to direct contact with *C. citratus* shows no evident breakage; however, the hyphae appear shriveled and wrinkled (Figure 6C). Above the MIC, the hyphae exhibit clear breakage and more pronounced shriveling, as indicated by the arrows in Figure 6E.

It is worth noting that the absence of Tween 80 in the culture medium in tests with volatiles may have led to the development of sporangia (as presented in the Supplementary Material in Figures S10 and S11, the control, and in the concentrations of MIC for both EO, 2 and 4C). Thus, it is possible to observe that exposure to concentrations above the MIC for both oils studied delayed or inhibited the development of sporangia.

P. expansum showed high mycelial density in all treatments exposed to volatiles and under direct contact (Supplementary

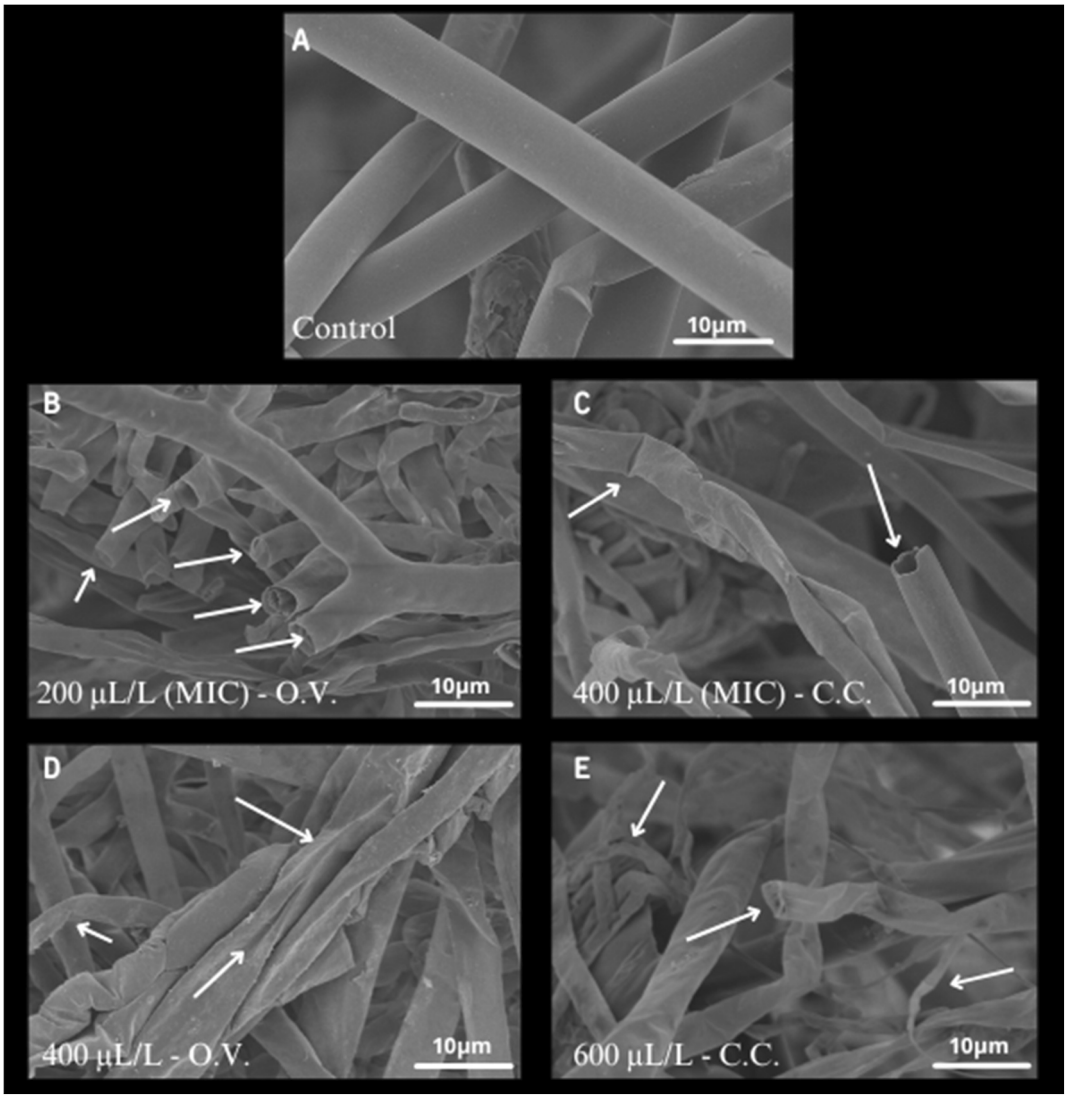

FIGURE 5 | SEM morphological observation of *Rhizopus stolonifer* exposed to volatile compounds from *Origanum vulgare* and *Cymbopogon citratus* essential oils. (A) Control—no exposure to essential oils (EOs). (B) Exposed to 200 µL/L $_{air}$ of *O. vulgare* EO (**MIC**). (C) Exposed to 400 µL/L $_{air}$ of *C. citratus* EO (**MIC**). (D) Exposed to 400 µL/L $_{air}$ of *O. vulgare* EO. (E) Exposed to 600 µL/L $_{air}$ of *C. citratus* EO. All images were captured at 1000× magnification.

Material S12 and S13—line 1), showing no clear differentiations at low magnitudes. The controls have intact conidiophore branches (metula, phialide, and conidium) (A in Figures 7 and 8); however, in the other treatments, changes in these structures are noticeable.

Under exposure to *O. vulgare* volatiles at the MIC concentration, it is possible to visualize the ruptured phialides, withered and twisted hyphae, and a few loose spores (Figure 7 - B). Above the MIC concentration, the reproductive structure becomes uncharacterized, and it also presents some loose spores, with hyphae showing tears/holes in the central region (Figure 7 - D). In the treatment with *C. citratus* volatiles, few spores are visualized in the images. Furthermore, at the MIC concentration, both perforations and phialides without conidia were observed (Figure 7 - C).

In direct contact, perforations in the hyphae and the mischaracterization of the sexual structure are observed for the two concentrations of *O. vulgare* (B and D in Figure 8). Furthermore, above the MIC, it is possible to observe a large amount of free spores surrounding the hyphae (Figure 8 - D). In contrast, the direct contact micrographs demonstrate no evident perforations in the hyphae, indicating the lower aggressiveness of *C. citratus* toward *P. expansum* (Figure 8 - C and E).

Kong et al. 2020 found inhibition and change in the morphology of *Colletotrichum gloeosporioides* when exposed to volatile organic compounds. In this study, the hyphae became rougher, with evident wrinkles and collapsed areas when in contact with volatiles rich in 2-phenylethyl methyl ether, phenylethyl alcohol, and 3-methyl-1-butanol produced by *Rahnella aquatilis* JZ-GX1 (bacteria isolated from the rhizosphere). Similar

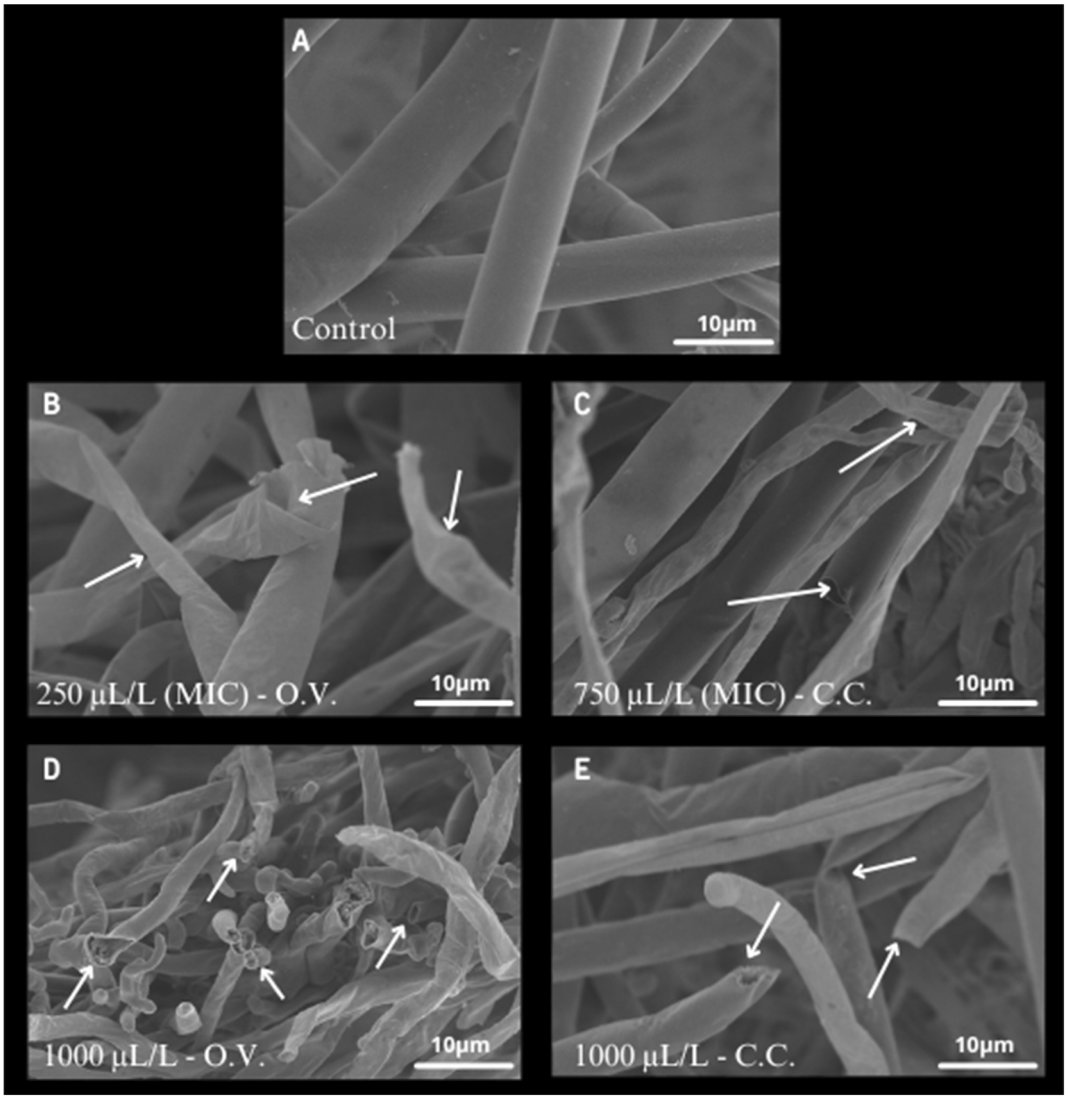

FIGURE 6 | **SEM morphological observation of *Rhizopus stolonifer* exposed to direct contact with *Origanum vulgare* and *Cymbopogon citratus* essential oils.** (A) Control—no exposure to essential oils (EOs). (B) Exposed to 250 µL/L of *O. vulgare* EO (**MIC**). (C) Exposed to 750 µL/L of *C. citratus* EO (**MIC**). (D) Exposed to 1000 µL/L of *O. vulgare* EO. (E) Exposed to 1000 µL/L of *C. citratus* EO. All images were captured at 1000× magnification.

changes in morphology were also observed in A*spergillus flavus* exposed to carvacrol (Duan et al. 2024) and exposed to *C. martinii* EO (Lin et al. 2023). The destruction of superficial wrinkles and distortions of the hyphae were observed in *R. stolonifer* exposed to a rich in thymol *L. sidoides* EO (Oliveira et al., 2019).

The main morphological changes observed in this work were changes in roughness and folds, breaks and holes, hollowing, and ribbon appearance in hyphae treated with *O. vulgare* and *C. citratus*. These changes strongly indicate structural changes in the cell wall and membrane, cytosol extravasation, and cell death. Duan et al. (2024) describe that carvacrol (the major component of *O. vulgare* EO) acts directly on the structural components of fungi, such as chitin and ergosterol. This compound is capable of interacting with and degrading chitin present in the cell wall and also acting in the gene regulation of chitinases. Ergosterol is an important component of the cell membrane responsible for the integrity and regulation of cell fluidity. Carvacrol works by interfering with the ergosterol synthesis cascade. Furthermore, citral and geraniol (major components of *C. citratus* EO) are related to the induction of necrotic and apoptotic cell death (Scariot et al. 2021). The images presented in this session improve the interpretation of data obtained in vitro tests with relevant phytopathogenic fungi in postharvest environments. They further highlight the potential utility of *O. vulgare* EO in combating fungal contamination.

EO, including those derived from *O. vulgare*, offer a sustainable approach to controlling postharvest diseases by attacking fungal pathogens directly through their structural components such as chitin and ergosterol. This approach not only mitigates the spread of fungal infections but also aligns with sustainable development goals by reducing food waste and contributing to food security. By offering an environmentally friendly alternative to conventional chemical treatments, EO help reduce dependence on synthetic fungicides, thereby supporting global efforts to minimize environmental impact and promote responsible

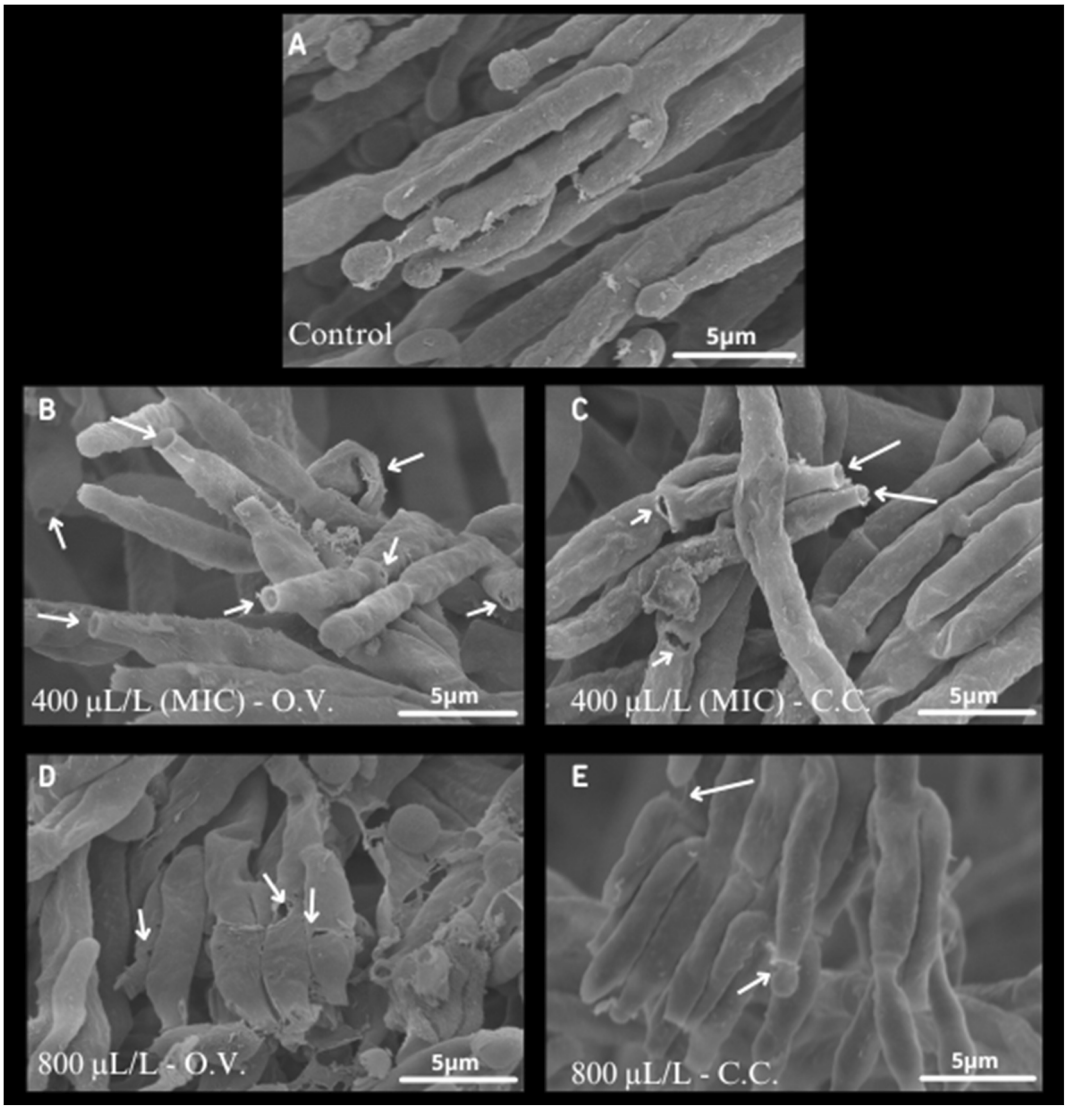

FIGURE 7 | **SEM morphological observation of *Penicillium expansum* exposed to volatile compounds from *Origanum vulgare* and *Cymbopogon citratus* essential oils.** (A) Control—no exposure to essential oils (EOs). (B) Exposed to 400 µL/L $_{air}$ of *O. vulgare* EO (**MIC**). (C) Exposed to 400 µL/L $_{air}$ of *C. citratus* EO (**MIC**). (D) Exposed to 800 µL/L $_{air}$ of *O. vulgare* EO. (E) Exposed to 800 µL/L $_{air}$ of *C. citratus* EO. All images were captured at 5000× magnification.

resource use. The visual evidence provided by these images affirms theoretical claims about the modes of action of oils and highlights their role in improving food preservation and reducing postharvest losses.

4 | Conclusion

Among EO tested in this study, *O. vulgare* L. was the most effective against the tested fungi. *Syzygium aromaticum* and *Cymbopogon citratus* also showed great antifungal activity using both methods, volatile exposure and direct contact. Considering these EO, *C. citratus* may be a great alternative to incorporate in postharvest fungal control, once it has a less intense odor that would interfere less with the sensory characteristics of fruits and vegetables. With the development of techniques that minimize the sensory perception of *O. vulgare* (such as nano and microencapsulation in inert matrices), this oil can also be used for this purpose. Overall, EO with alcohol major compounds tended to have lower MIC than other EO, showing great potential for fungal control.

The volatile exposure and direct contact methods demonstrated varying levels of effectiveness depending on the fungal species (*Colletotrichum* sp. and *Rhizopus stolonifer* exhibited greater susceptibility to the effects of exposure to volatiles from *O. vulgare* and *M. piperita*, respectively, whereas *Penicillium expansum* showed higher sensitivity to the effects of direct contact with *O. vulgare*), indicating that the most effective control strategy may differ based on the specific microorganism. These findings also highlight the potential possibility of *O. vulgare* L., *S. aromaticum*, and *C. citratus* EO as antifungal agents for incorporation into edible coatings (direct contact) and

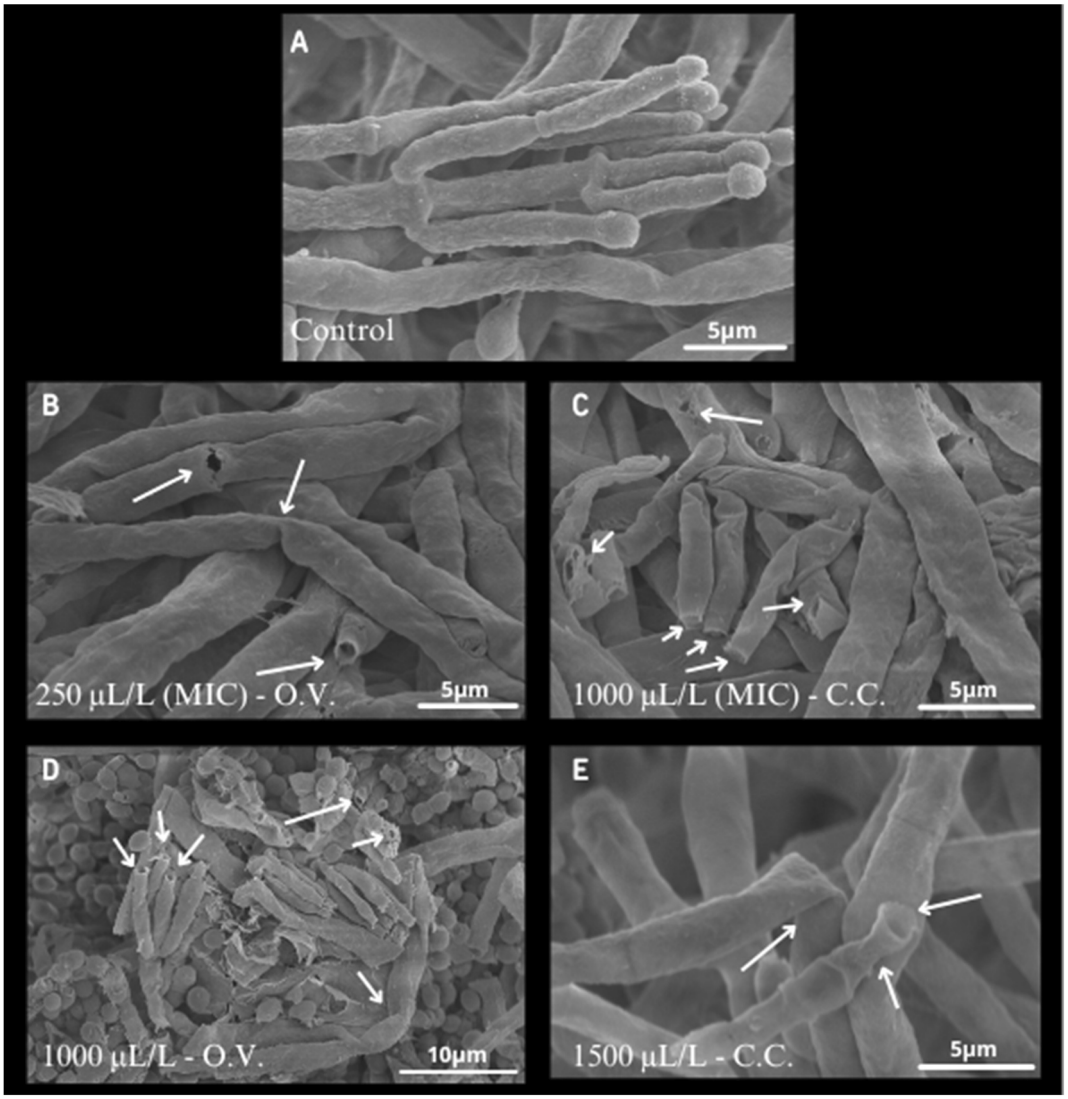

FIGURE 8 | SEM morphological observation of *Penicillium expansum* exposed to direct contact with *Origanum vulgare* and *Cymbopogon citratus* essential oils. (A) Control—no exposure to essential oils (EOs). (B) Exposed to 250 µL/L of *O. vulgare* EO (**MIC**). (C) Exposed to 1000 µL/L of *C. citratus* EO (**MIC**). (D) Exposed to 1000 µL/L of *O. vulgare* EO. (E) Exposed to 1500 µL/L of *C. citratus* EO. Images A, B, C, and E were captured at 5000× magnification, whereas image D was captured at 1000× magnification to highlight the concentration of spores surrounding the hypha.

active packaging (volatile application) in future investigations aiming to extend the shelf life of fruits and vegetables.

In addition, the scanning electron microscopy images improve the interpretation of data obtained from in vitro tests with *Colletotrichum* sp., *Rhizopus stolonifer,* and *Penicillium expansum*. The observation of changes (roughness and folds, breaks and holes, hollowing, and ribbon appearance) in hyphae contributed to visually confirming the mechanisms of cellular action of the essential oils of *O. vulgare* and *C. citratus* as described in the literature.

Author Contributions

Conny W.T. Fukuyama: formal analysis, investigation, writing – original draft. **Ramon Peres Brexó:** formal analysis, investigation, writing – review and editing. **Larissa G.R. Duarte:** methodology, writing – review and editing. **Maria Eduarda M. Martins:** investigation. **Maria Eduarda A. Astolfo:** investigation. **Ygor G.P. Osti:** investigation. **Isadora C. Pedrino:** Investigation. **Higor V. Santos:** investigation. **Josemar de Oliveira Filho:** investigation. **Fernanda Ramalho Procopio:** writing – review and editing. **Stanislau Bogusz Junior:** methodology, project administration. **Marcos D. Ferreira:** conceptualization, funding acquisition, methodology, project administration, resources, supervision, writing – review and editing.

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opinions, hypotheses, and conclusions or recommendations expressed in this article are the responsibility of the author(s) and do not necessarily reflect the view of FAPESP.

Conflicts of Interest

The authors declare no conflicts of interest.

Data Availability Statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

References

Abdi-Moghadam, Z., Y. Mazaheri, A. Rezagholizade-shirvan, et al. 2023. "The Significance of Essential Oils and Their Antifungal Properties in the Food Industry: A Systematic Review." *Heliyon* 9, no. 11: e21386. https://doi.org/10.1016/j.heliyon.2023.e21386.

Achimón, F., V. D. Brito, R. P. Pizzolitto, A. Ramirez Sanchez, E. A. Gómez, and J. A. Zygadlo. 2021. "Chemical Composition and Antifungal Properties of Commercial Essential Oils Against the Maize Phytopathogenic Fungus Fusarium Verticillioides." *Revista Argentina de Microbiología* 53, no. 4: 292–303. https://doi.org/10.1016/j.ram.2020.12.001.

Aguilar-González, A. E., E. Palou, and A. López-Malo. 2015. "Antifungal Activity of Essential Oils of Clove (Syzygium Aromaticum) And/or Mustard (Brassica Nigra) in Vapor Phase Against Gray Mold (Botrytis Cinerea) in Strawberries." *Innovative Food Science & Emerging Technologies* 32: 181–185. https://doi.org/10.1016/j.ifset.2015.09.003.

Al-Maqtari, Q. A., A. Rehman, A. A. Mahdi, et al. 2022. "Application of Essential Oils as Preservatives in Food Systems: Challenges and Future Perspectives – a Review." *Phytochemistry Reviews* 21, no. 4: 1209–1246. https://doi.org/10.1007/s11101-021-09776-y.

Almeida, N. A., L. Freire, L. Carnielli-Queiroz, A. P. A. Bragotto, N. C. C. Silva, and L. O. Rocha. 2024. "Essential Oils: An Eco-friendly Alternative for Controlling Toxigenic Fungi in Cereal Grains." *Comprehensive Reviews in Food Science and Food Safety* 23, no. 1. https://doi.org/10.1111/1541-4337.13251.

Álvarez-García, S., M. Moumni, and G. Romanazzi. 2023. "Antifungal Activity of Volatile Organic Compounds From Essential Oils Against the Postharvest Pathogens Botrytis Cinerea, Monilinia Fructicola, Monilinia Fructigena, and Monilinia Laxa." *Frontiers in Plant Science* 14. https://doi.org/10.3389/fpls.2023.1274770.

Andrade-Ochoa, S., K. F. Chacón-Vargas, L. E. Sánchez-Torres, B. E. Rivera-Chavira, B. Nogueda-Torres, and G. V. Nevárez-Moorillón. 2021. "Differential Antimicrobial Effect of Essential Oils and Their Main Components: Insights Based on the Cell Membrane and External Structure." *Membranes* 11, no. 6: 405. https://doi.org/10.3390/membranes11060405.

Bertolo, M. R. V., J. G. de Oliveira Filho, G. C. Lamonica, et al. 2025. "Improvement of the Physical-Chemical, Microbiological, Volatiles and Sensory Quality of Strawberries Covered With Chitosan/gelatin/pomegranate Peel Extract-Based Coatings." *Food Chemistry* 471: 142755. https://doi.org/10.1016/j.foodchem.2025.142755.

Bouyahya, A., H. Mechchate, T. Benali, et al. 2021. "Health Benefits and Pharmacological Properties of Carvone." *Biomolecules* 11, no. 12: 1803. https://doi.org/10.3390/biom11121803.

Carmona-Hernandez, S., J. Reyes-Pérez, R. Chiquito-Contreras, G. Rincon-Enriquez, C. Cerdan-Cabrera, and L. Hernandez-Montiel. 2019. "Biocontrol of Postharvest Fruit Fungal Diseases by Bacterial Antagonists: A Review." *Agronomy* 9, no. 3: 121. https://doi.org/10.3390/agronomy9030121.

Chizzola, R. 2013. "Regular Monoterpenes and Sesquiterpenes (Essential Oils)." In *Natural Products: Phytochemistry, Botany and Metabolism of Alkaloids, Phenolics and Terpenes*, edited by K. Ramawat, J. M. Mérillon, 2973–3008. Springer. https://doi.org/10.1007/978-3-642-22144-6_130.

Devi, M. A., D. Sahoo, T. B. Singh, and Y. Rajashekar. 2021. "Antifungal Activity and Volatile Organic Compounds Analysis of Essential Oils From Cymbopogon Species Using Solid-phase Microextraction-Gas Chromatography-Mass Spectrometry." *Journal of Agriculture and Food Research* 3: 100110. https://doi.org/10.1016/j.jafr.2021.100110.

Dora, M., S. Biswas, S. Choudhary, R. Nayak, and Z. Irani. 2021. "A System-wide Interdisciplinary Conceptual Framework for Food Loss and Waste Mitigation Strategies in the Supply Chain." *Industrial Marketing Management* 93: 492–508. https://doi.org/10.1016/j.indmarman.2020.10.013.

Duan, W.-Y., X.-M. Zhu, S.-B. Zhang, et al. 2024. "Antifungal Effects of Carvacrol, the Main Volatile Compound in Origanum Vulgare L. Essential Oil, Against Aspergillus Flavus in Postharvest Wheat." *International Journal of Food Microbiology* 410: 110514. https://doi.org/10.1016/j.ijfoodmicro.2023.110514.

FAO. 2015. "Global Initiative on Food Loss and Waste Reduction." *Food and Agriculture Organization of the United Nations* 1–8. https://www.fao.org/3/i7657e/i7657e.pdf.

Fukuyama, C. W. T., L. G. R. Duarte, I. C. Pedrino, M. C. Mitsuyuki, S. B. Junior, and M. D. Ferreira. 2024. "Effect of Carnauba Wax Nanoemulsion Associated With Syzygium Aromaticum and Mentha Piperita Essential Oils as an Alternative to Extend Lychee Post-harvest Shelf Life." *Sustainable Food Technology* 2: 426–436. https://doi.org/10.1039/D3FB00251A.

Galsurker, O., S. Diskin, D. Maurer, O. Feygenberg, and N. Alkan. 2018. "Fruit Stem-End Rot." *Horticulturae* 4: 50. https://doi.org/10.3390/horticulturae4040050.

Giménez-Santamarina, S., J. A. Llorens-Molina, F. Sempere-Ferre, C. Santamarina, J. Roselló, and M. P. Santamarina. 2022. "Chemical Composition of Essential Oils of Three Mentha Species and Their Antifungal Activity Against Selected Phytopathogenic and Post-harvest Fungi." *All Life* 15, no. 1: 64–73. https://doi.org/10.1080/26895293.2021.2022007.

Hudz, N., L. Kobylinska, K. Pokajewicz, et al. 2023. "Mentha Piperita: Essential Oil and Extracts, Their Biological Activities, and Perspectives on the Development of New Medicinal and Cosmetic Products." *Molecules* 28, no. 21: 7444. https://doi.org/10.3390/molecules28217444.

Jijakli, M. H., and P. Lepoivre. 2004. "State of the Art and Challenges of Post-harvest Disease Management in Apples." In *Fruit and Vegetable Diseases*, Vol. 1 edited by K. G. Mukerji, 59–94. Kluwer Academic Publishers, Springer. https://doi.org/10.1007/0-306-48575-3_3.

Karoney, E. M., T. Molelekoa, M. Bill, N. Siyoum, and L. Korsten. 2024. "Global Research Network Analysis of Fresh Produce Postharvest Technology: Innovative Trends for Loss Reduction." *Postharvest Biology and Technology* 208: 112642. https://doi.org/10.1016/j.postharvbio.2023.112642.

Kong, W.-L., L. Rui, H. Ni, and X.-Q. Wu. 2020. "Antifungal Effects of Volatile Organic Compounds Produced by Rahnella Aquatilis JZ-GX1 Against Colletotrichum Gloeosporioides in Liriodendron Chinense × Tulipifera." *Frontiers in Microbiology* 11. https://doi.org/10.3389/fmicb.2020.01114.

Lee, J.-E., S.-M. Seo, M.-J. Huh, S.-C. Lee, and I.-K. Park. 2020. "Reactive Oxygen Species Mediated-Antifungal Activity of Cinnamon Bark (Cinnamomum Verum) and Lemongrass (Cymbopogon Citratus) Essential Oils and Their Constituents Against Two Phytopathogenic Fungi." *Pesticide Biochemistry and Physiology* 168: 104644. https://doi.org/10.1016/j.pestbp.2020.104644.

Leonelli Pires de Campos, A. C., R. D. Saldanha Nandi, S. Scandorieiro, et al. 2022. "Antimicrobial Effect of Origanum Vulgare (L.) Essential Oil as an Alternative for Conventional Additives in the Minas Cheese

Manufacture." *Lebensmittel-Wissenschaft & Technologie* 157: 113063. https://doi.org/10.1016/j.lwt.2021.113063.

Lin, L., S. Chen, W. Xia, et al. 2023. "A New Strategy: Inhibitory Effect and Mechanism of Cymbopogon Martinii Essential Oil on Aspergillus flavus." *Food Bioscience* 51: 102315. https://doi.org/10.1016/j.fbio.2022.102315.

Madi, Y. F., M. A. Choucry, M. R. Meselhy, and E. S. A. El-Kashoury. 2021. "Essential Oil of Cymbopogon Citratus Cultivated in Egypt: Seasonal Variation in Chemical Composition and Anticholinesterase Activity." *Natural Product Research* 35: 4063–4067. https://doi.org/10.1080/14786419.2019.1644320.

Mansoori, S., H. Bahmanyar, E. Jafari Ozumchelouei, and I. Najafipour. 2022. "Investigation and Optimisation of the Extraction of Carvone and Limonene From the Iranian Mentha Spicata through the Ultrasound-Assisted Extraction Method." *Indian Chemical Engineer* 64, no. 2: 141–150. https://doi.org/10.1080/00194506.2020.1831407.

Maurya, A., J. Prasad, S. Das, and A. K. Dwivedy. 2021. "Essential Oils and Their Application in Food Safety." *Frontiers in Sustainable Food Systems* 5. https://doi.org/10.3389/fsufs.2021.653420.

Nunes, C. S., and P. Philipps-Wiemann. 2018. "Chitinases." In *Enzymes in Human and Animal Nutrition*, edited by B. W. T. Hardy and J. L. Gilliland, 361–378. Elsevier. https://doi.org/10.1016/B978-0-12-805419-2.00018-6.

Nybom, H., M. Ahmadi-Afzadi, K. Rumpunen, and I. Tahir. 2020. "Review of the Impact of Apple Fruit Ripening, Texture and Chemical Contents on Genetically Determined Susceptibility to Storage Rots." *Plants* 9, no. 7: 831. https://doi.org/10.3390/plants9070831.

Oliveira Filho, J. G., G. da Cruz Silva, A. C. de Aguiar, et al. 2021. "Chemical Composition and Antifungal Activity of Essential Oils and Their Combinations Against Botrytis Cinerea in Strawberries." *Journal of Food Measurement and Characterization* 15, no. 2: 1815–1825. https://doi.org/10.1007/s11694-020-00765-x.

Plata-Rueda, A., L. C. Martínez, G. S. Rolim, et al. 2020. "Insecticidal and Repellent Activities of Cymbopogon Citratus (Poaceae) Essential Oil and its Terpenoids (Citral and Geranyl Acetate) Against Ulomoides Dermestoides." *Crop Protection* 137: 105299. https://doi.org/10.1016/j.cropro.2020.105299.

Reyes-Jurado, F., A. R. Navarro-Cruz, C. E. Ochoa-Velasco, E. Palou, A. López-Malo, and R. Ávila-Sosa. 2020. "Essential Oils in Vapor Phase as Alternative Antimicrobials: A Review." *Critical Reviews in Food Science and Nutrition* 60, no. 10: 1641–1650. https://doi.org/10.1080/10408398.2019.1586641.

Saleh, I., and R. Al-Thani. 2019. "Fungal Food Spoilage of Supermarkets' Displayed Fruits." *Veterinary World* 12, no. 11: 1877–1883. https://doi.org/10.14202/vetworld.2019.1877-1883.

Scariot, F. J., M. S. Pansera, A. P. L. Delamare, and S. Echeverrigaray. 2021. "Citral and Geraniol Induce Necrotic and Apoptotic Cell Death on Saccharomyces cerevisiae." *World Journal of Microbiology and Biotechnology* 37: 3. https://doi.org/10.1007/s11274-021-03011-8.

Sen, S., M. Israr, S. Singh, M. K. Singh, R. S. Verma, and D. U. Bawankule. 2023. "Pharmaceutical, Cosmeceutical, Food Additive and Agricultural Perspectives of Cymbopogon Martini: A Potential Industrial Aromatic Crop." *South African Journal of Botany* 158: 277–291. https://doi.org/10.1016/j.sajb.2023.05.007.

Soliman, S. A., E. E. Hafez, A. M. G. Al-Kolaibe, et al. 2022. "Biochemical Characterization, Antifungal Activity, and Relative Gene Expression of Two Mentha Essential Oils Controlling Fusarium Oxysporum, the Causal Agent of Lycopersicon esculentum Root Rot." *Plants* 11, no. 2: 189. https://doi.org/10.3390/plants11020189.

Spencer, P. V. D., S. H. Libardi, F. F. G. Dias, et al. 2021. "Chemical Composition, Antioxidant and Antibacterial Activities of Essential Oil From Cymbopogon Densiflorus (Steud.) Stapf Flowers." *Journal of Essential Oil Bearing Plants* 24, no. 1: 40–52. https://doi.org/10.1080/0972060X.2020.1862711.

Tullio, V., A. Nostro, N. Mandras, et al. 2007. "Antifungal Activity of Essential Oils Against Filamentous Fungi Determined by Broth Microdilution and Vapour Contact Methods." *Journal of Applied Microbiology* 102, no. 6: 1544–1550. https://doi.org/10.1111/j.1365-2672.2006.03191.x.

Ventura-Aguilar, R. I., E. P. Díaz-Galindo, S. Bautista-Baños, et al. 2021. "Monitoring the Infection Process of Rhizopus Stolonifer on Strawberry Fruit During Storage Using Films Based on Chitosan/polyvinyl Alcohol/polyvinylpyrrolidone and Plant Extracts." *International Journal of Biological Macromolecules* 182: 583–594. https://doi.org/10.1016/j.ijbiomac.2021.03.187.

Yu, H., Z.-X. Lin, W.-L. Xiang, et al. 2022. "Antifungal Activity and Mechanism of D-Limonene Against Foodborne Opportunistic Pathogen Candida Tropicalis." *Lebensmittel-Wissenschaft & Technologie* 159: 113144. https://doi.org/10.1016/j.lwt.2022.113144.

Zang, E., L. Jiang, H. Cui, et al. 2023. "Only Plant-Based Food Additives: An Overview on Application, Safety, and Key Challenges in the Food Industry." *Food Reviews International* 39, no. 8: 5132–5163. https://doi.org/10.1080/87559129.2022.2062764.

Zhao, P., J. P. Ndayambaje, X. Liu, and X. Xia. 2022. "Microbial Spoilage of Fruits: A Review on Causes and Prevention Methods." *Food Reviews International* 38, no. sup1: 225–246. https://doi.org/10.1080/87559129.2020.185885.

Supporting Information

Additional supporting information can be found online in the Supporting Information section.