



Essential oils of oregano and cinnamon as an alternative method for control of gray mold disease of table grapes caused by *Botrytis cinerea*

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Abstract

The study examined the potential of essential oils for controlling gray mold disease of table grape caused by *Botrytis cinerea*. Essential oils of cinnamon and oregano were used in vitro and in vivo. Both oils reduced the mycelial growth of the pathogen at various concentrations (100, 200, 400, 600, 800, and 1000 µl per liter). The most effective concentrations of both essential oils were 800 and 1000 µL/L, which yielded the highest reduction in pathogen growth. Both oils reduced the spore germination of the pathogen at 800 µL/L. In addition, cinnamon and oregano oils reduced the disease severity of gray mold by 58.9% and 42%, respectively. Each essential oil was able to increase peroxidase and polyphenol oxidase enzymes and phenol and flavonoid content in table grapes infected with *B. cinerea* compared with the control. Interestingly, essential oils increased total soluble solids, vitamin C (VC), and titratable acidity (TS) in fruits compared to the control. In conclusion, our study confirmed that cinnamon and oregano may be applied as future ecofriendly alternatives to synthetic fungicides for controlling gray mold disease.

Keywords Antioxidant · Disease reduction · Enzyme activities · Spore germination · Titratable acidity · Total soluble solids · Vitamin C

Introduction

Botrytis cinerea Pers. Fr. (teleomorph *Botryotinia fuckeliana* [de Bary] Whetzel) is the most significant pathogen of table grapes in many countries (Simone et al. 2020). Gray mold disease caused by *B. cinerea* can affect a wide range of fresh fruits and vegetables around the world, including table grapes, resulting in significant production and profit losses (Youssef et al. 2020). The most common treatment for this disease in grapes and other horticultural crops is fungicides. However, these have been limited in their application due to various factors such as legislation, resistance, human health, and the environment (Bagy et al. 2021).

Many researchers have searched for new disease control methods that are non-toxic. Several alternative methods have been proposed to control postharvest diseases of vegetables and fruit. Plant extracts (Chen et al. 2019), controlled atmosphere (Santana et al. 2011), heat (Wilson-Wijeratnam et al. 2005), postharvest salicylic acid or chitosan (Bagy et al. 2021; Katiyar et al. 2015), biological control (Eshel et al. 2009; Abo-Elyousr et al. 2021), and essential oils (Boubaker et al. 2016; Rosa Vilaplana et al. 2018) have been investigated, and these methods can be effective against pathogens but have less effect on non-pathogenic organisms (Sallam et al. 2012; Youssef et al. 2019). Essential oils (EOs) are one type of alternative control strategy, and are known for their antimicrobial and biodegradable properties as well as their lack of residual effect on fresh produce (Bakkali et al. 2008). They improve the quality of the fruit and lengthen its shelf life (Plaza et al. 2004) and antioxidant properties (Sivakumar and Bautista-Baños 2014). EOs may be a promising advancement in the battle against fruit postharvest decay by reducing the necessity of chemical fungicide handling (Maqbool et al. 2010). These oils

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inhibit pathogens because they have a direct effect on mycelial growth and spore germination by changing the microorganism's cellular metabolism. (Sivakumar and Bautista-Baños 2014). Using natural products derived from plant extracts can potentially alter fruit sensory quality. Fruit acceptability can suffer as a result of off-flavor and changes in aroma and flavor brought on by the strong odor of EOs (Perdones et al. 2012; Sangsuwan et al. 2016), but at low concentrations, these limitations are not of great importance (Vilaplana et al. 2018).

Treatment of plants with various bioagents can protect against pathogens by inducing local and systemic resistances (Abo-Elyousr et al. 2021). Application of some postharvest treatments increases the antioxidant mechanism in plants, which increases their antioxidant levels (Youssef et al. 2020), including antioxidant enzymes PPO and POD (Ballester et al. 2006). Nonenzymatic antioxidants are the other side of the antioxidant mechanism, and include phenolics, flavonoids, and others (de Pinto and De Gara 2004).

The main objective of this study was to evaluate the effect of cinnamon and oregano oils on reduction of gray mold disease caused by *B. cinerea* and the changes in activity of some enzymes, e.g., peroxidase (POD) and PPO, as well as total phenolic content and total flavonoid content in grape tissues along with their physiochemical changes.

Materials and methods

Source of plant EOs

EOs of cinnamon (*Cinnamomum verum* L.; main volatile compounds: oxygenated terpenoids and terpene hydrocarbons α -copaene and α -bergamotene) and oregano (*Origanum vulgare* L.; main volatile compounds: eugenol, 2-phenyl-ethanol, thymoquinone benzyl alcohol, thymol, 3-hexen-1-ol, and carvacrol) were used in this study for purity. EOs (100%) were obtained from the EL-Masrayia natural oils company, Egypt. These EOs were stored in dark bottles at 4 °C until use in the experiments. Information about volatile compounds was provided by EL-Masrayia.

Plant materials

Table grape (*Vitis vinifera* L.) cv. “Taify” fruits were harvested from a private orchard in the Taif region (Saudi Arabia), transported to the laboratory, and put into cold rooms for precooling at 1 °C \pm 2 °C. Bunches symmetrical in size and free from pathological symptoms or mechanical damage were used in the experiment. Fruits were randomly selected for the treatments.

Isolation and molecular identification of gray mold pathogen

Botrytis cinerea was isolated from grape fruits showing typical symptoms of gray mold disease on Potato Dextrose Agar (PDA) medium. The pathogen was morphologically identified based on colony color, mycelial growth, and spore shape as described by Domsch et al. (1980). Molecular identification of the pathogen was confirmed by the 18S ribosomal DNA gene. Obtained sequences were compared with the sequences available in public domain of National Center for Biotechnology Information (NCBI) library using Basic Local Alignment Search Tool (BLAST). Products were identified based on the higher similarity and sequences were submitted to NCBI under specific accession numbers. Phylogenetic tree was constructed with ITS1 sequences by neighbor joining algorithm in MEGA 6X package (Tamura et al. 2013).

In vitro antifungal assay

Antifungal activity of essential oil against mycelial growth of the pathogen

The antifungal activity of cinnamon (CV) and oregano (OV) EOs was assessed on *B. cinerea*. Using the produced stock solutions, the EOs were applied to conical flasks containing 100 mL of sterile PDA medium before solidification to obtain the required concentrations of 100, 200, 400, 600, 800, and 1000 μ L/L. Nearly 20 mL of enriched media was poured into each Petri plate (9 cm). Discs (5 mm diameter) of pathogen were cultured for 10 d at 25 °C–28 °C. Reduction in mycelial growth was calculated relative to the growth in the control treatment. Eight plates were used for each concentration. Fungal growth was recorded after 10 d by measurement of the orthogonal diameter using the following formula:

$$I\% = (A - B/A) \times 100$$

where I% is the percent mycelial growth inhibition, A is the growth diameter of the fungi in the control, and B is the growth diameter (cm) of the pathogen for each concentration of the essential oil. Each experiment was performed twice, with eight plates per concentration. The concentrations that significantly reduced *B. cinerea* mycelial growth were selected for additional experiments.

Effects on spore germination

The effects of two EOs at a concentration of 800 μ L/L on spore germination were determined. The EOs at

concentration of 800 was added to water agar medium. Conidia germination was observed on 1.5% water agar medium plates. A total of 1 mL of spore suspension (1×10^6 conidia/mL) from each treatment was spread with a sterilized glass spreader over water agar (agar 20 g; distilled water, 1000 mL) plates. Plates were incubated for 12 h at 22 °C. The number of germinated conidia was counted in each plate. Conidia were said to be germinated if the germ tube was longer than the conidia. Germinated conidia were expressed as percentage germination as follows:

$$\text{number of germinated conidia} / \text{total number of conidia} \times 100$$

Three replicates were used, and the experiment was conducted twice, as described by Elsherbiny et al. (2021).

Activity of essential oil of cinnamon and oregano EOs in vivo

Preparation of pathogen inoculum

Botrytis cinerea was transferred onto PDA plates and incubated at 25 °C for 10 d to prepare the inoculum. After inoculation, each plate received 10 mL distilled water, and the spores were scraped into distilled water with a sterilized bacterial L-shape rod. After that, the spore suspension was passed through a sterile muslin sheet. The concentration of the conidial suspension was adjusted to 1×10^6 conidia/mL using a hemocytometer (Abdel-Rahim and Abo-Elyousr 2018).

Effects of cinnamon and oregano EOs on gray mold disease of grape fruits

The effects of cinnamon and oregano EOs on grape fruits were examined using the protocols described by Youssef et al. (2020), with minor modifications. Briefly, healthy table grape (*Vitis vinifera* L.) cv. “Taify” fruits were purchased, surface sterilized with 2% sodium hypochlorite for 2 min, washed with tap water three times, and air-dried at room temperature. According to the method of Pedrotti et al. (2017), the antifungal activity of both EOs on grapes was evaluated. From each grape cluster, 10 berries were wounded to approximately 2 mm depth. After wounding, a conidial suspension of *B. cinerea* at 1×10^6 conidia/mL was inoculated (10 µL in each wound). After 4 h, grape bunches were sprayed with the most effective concentration of the oils at 800 µL/L from both oils till run-off. Treated berries were placed in plastic boxes (20 × 13 × 10 cm) and incubated at 2 °C ± 1 °C and high humidity (90%–95%) with a 16 h photoperiod of 8 d for those inoculated with *B. cinerea*. After incubation, disease severity was assessed. For each incidence, 10 inoculated berries from each cluster of grapes

were evaluated, and disease severity was visually evaluated using a scale from 0 to 100% as described previously (Pedrotti et al. 2017; Madbouly et al. 2020).

Effects of cinnamon and oregano EOs on peroxidase (POD) and polyphenol oxidase (PPO) activity in grape tissues after inoculation with the pathogen

Enzyme extraction

To extract enzymes from the grapes, 200 mg lyophilized grape tissues were homogenized with 20 mM Tris–HCl buffer (pH 7.2) using a homogenizer. The homogenate was placed in glass tubes and centrifuged at 4 °C for 10 min at 10,000 rpm. The supernatant was then labeled as crude extract and stored at –20 °C until PPO and POD activity were determined.

POD and PPO activity

The method of Putter (1974) was used to measure POD activity. Each prepared reaction mixture comprised 1 mL in total and contained 8 µL (0.97 M) H₂O₂, 0.25 mL (0.2 M) sodium acetate (pH 5.5), 0.08 mL (0.5 M) guaiacol, and the appropriate amount of enzyme preparation. Extraction buffer alone served as a blank reference sample. Three replicates were used for each treatment. PPO activity was determined following protocols from Batra and Kuhn (1975), using catechol as a substrate.

Nonenzymatic assays

Sample preparation For phenolic and flavonoid compound extraction, 1.0 g sample was suspended in 10 mL 70% ethanol (v/v). The suspension was then placed at 30 °C on a shaker at 120 rpm for 2 h and then centrifuged at 1013 × g for 5 min. The resulting supernatant was used for further analysis.

Total phenol and flavonoid content The total phenol content of each sample was measured according to Malik and Singh (1980). The standard curve was prepared using gallic acid, and the phenol content in each extract was expressed as mg gallic acid/g fresh weight.

Flavonoid concentrations in each sample were determined using the colorimetric assay reported by Zhishen et al. (1999). Known concentrations of catechin were used to create a standard curve, and the results were described as mg catechin equivalent/g.

Effects of cinnamon and oregano EOs on chemical properties of fruits

Total soluble solids

A digital refractometer (Pocket Refractometer PAL 3, ATAGO, Japan) was used to determine the percentage of total soluble solids (TSS) in the fruit juice according to the A.O.A.C. (2000)

Titrateable acidity (%) and vitamin C

Titrateable acidity was determined by potentiometric titration with 0.1 N NaOH to pH 8.2, using 10 mL of diluted juice in 40 mL distilled water. The results were expressed as tartaric acid (%). Vitamin C value was estimated according to the A.O.A.C. (2000) and presented as mg/100 mL of juice.

Statistical analysis

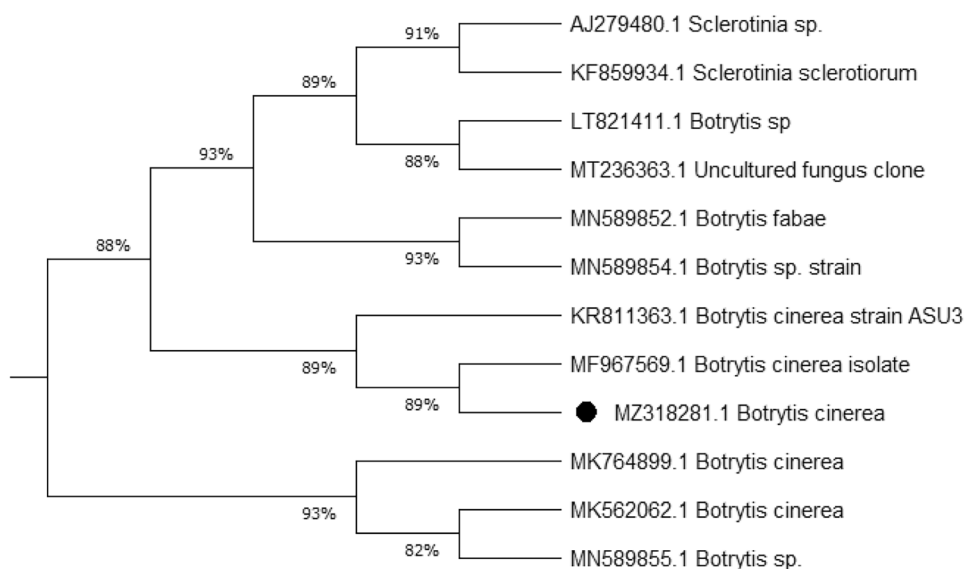
Experimental data were analyzed using via one-way ANOVA in the MSTATC program. On the other hand, results of the enzymes, non-enzyme activities, and physiochemical studies were analyzed based on two-way ANOVA. A least significant difference test at $P=0.05$ was performed to identify significant differences between the means of results of various treatments.

Results

Molecular identification

Botrytis cinerea strain NAMK7 was isolated from gray mold rot disease of grape table in this study. Based on

Fig. 1 Phylogenetic analysis of *Botrytis cinerea* isolate on the ITS gene. This analysis was performed using the neighbor-joining method in BLAST pairwise alignments



its culture and microscopic characteristics, the isolated fungus was identified as *B. cinerea*. The strain NAMK7 has a 100% similarity resemblance to *B. cinerea* strain ASU3 (KR811363), according to an amplified and sequenced fragment of the 18S ribosomal DNA gene (491 base pairs). The *B. cinerea* strain NAMK7 sequence was deposited in the GenBank nucleotide sequence database under accession number MZ318281. The sequence alignment of the 18S rDNA gene was used to create a phylogenetic tree (Fig. 1) using MEGAX.

Antifungal activity of essential oil on *B. cinerea*

Mycelial growth

The inhibition rate of various concentrations of cinnamon and oregano EOs (100, 200, 400, 600, 800, and 1000 $\mu\text{L/L}$) against *B. cinerea* is shown in Table 1. The cinnamon and oregano EOs displayed high fungitoxicity toward *B. cinerea*, with decreased mycelial growth as the essential oil dose was increased. The minimum inhibitory concentrations of each oil for *B. cinerea* were 100 and 200 $\mu\text{L/L}$, whereas 400, 600 $\mu\text{L/L}$ showed suppression of pathogen mycelial growth. The most effective concentrations of both EOs for inhibition of *B. cinerea* growth were 800 and 1000 $\mu\text{L/L}$.

Spore germination

EOs (cinnamon and oregano) at 800 $\mu\text{L/L}$ showed significant activity against spore germination of *B. cinerea* compared with the control (Table 2). They decreased spore germination by approximately 65.2%.

Effects of cinnamon and oregano EOs on gray mold disease of table grape fruits (*Vitis vinifera* L.) cv. “Taify” in vivo

For this part of the study, cinnamon and oregano EOs at 800 $\mu\text{L/L}$ were tested in vivo to study their effect on the disease severity of gray mold disease of table grape “Taify” fruits (Table 3). Treatment of fruits with cinnamon or oregano oil significantly reduced the percentage of infected grape fruits compared to the control. Oregano oil yielded a higher reduction in the percentage of fruit infected with *B. cinerea* (39%) compared to cinnamon oil (55%). In addition, oregano oil was able to suppress the disease severity of gray mold more effectively than cinnamon oil.

Effects of cinnamon and oregano EOs on:

Peroxidase (POD) and polyphenol oxidase (PPO) activity

Two days after treatment of infected “Taify” grape fruits with each essential oil at 800 $\mu\text{L/L}$, the activity of the POD was increased compared to the control (Fig. 2). The cinnamon oil showed higher POD activity than oregano oil. The maximum level of PPO was visible at four days after treatment of fruit with EOs. The activity of POD was decreased at 6 days in all treatments.

Treatment of infected grape fruits with cinnamon and oregano oil increased PPO activity relative to the control

(Fig. 3). Two days after treatment of fruit with each essential oil, the enzyme activity began to increase. It peaked at 4 days, and then began to decline at 6 days. Overall, fruits treated with cinnamon oil showed higher PPO activity than those treated with oregano oil.

Total phenol and flavonoid content

The total phenol content was estimated at several time periods after treating infected “Taify” grape fruits with cinnamon and oregano oil at 800 $\mu\text{L/L}$ (Fig. 4). The highest total phenol content was found in fruits treated with cinnamon oil, followed by those treated with oregano oil. Fruits treated with cinnamon oil showed increased total phenol content at 2 days and remained stable at 4 days, while fruits treated with oregano reached the maximum at 4 days. After 6 days, the total phenol content was decreased.

The results showed that EOs were able to increase total flavonoid content in infected grape cv. “Taify” fruits on days 2–6 as compared to control fruits (Fig. 5). At 2 days, no significant increase was found in total flavonoid content in fruits treated with cinnamon and oregano oil. After 2 days, there was no significant difference in total flavonoid content between cinnamon and oregano oil. The cinnamon oil increased total flavonoid content at 4 days, whereas oregano oil decreased it. Both oils reduced the total flavonoid content at 6 days after treatment.

Fig. 2 Effect of treatments with essential oils of cinnamon (*Cinnamomum verum* L.) and oregano (*Origanum vulgare* L.) on peroxidase activity (U/min/g fw) in extracts of “Taify” grape tissues. Values in the column followed by different letters indicate significant differences among treatments according to a least significant differences test ($P=0.05$)

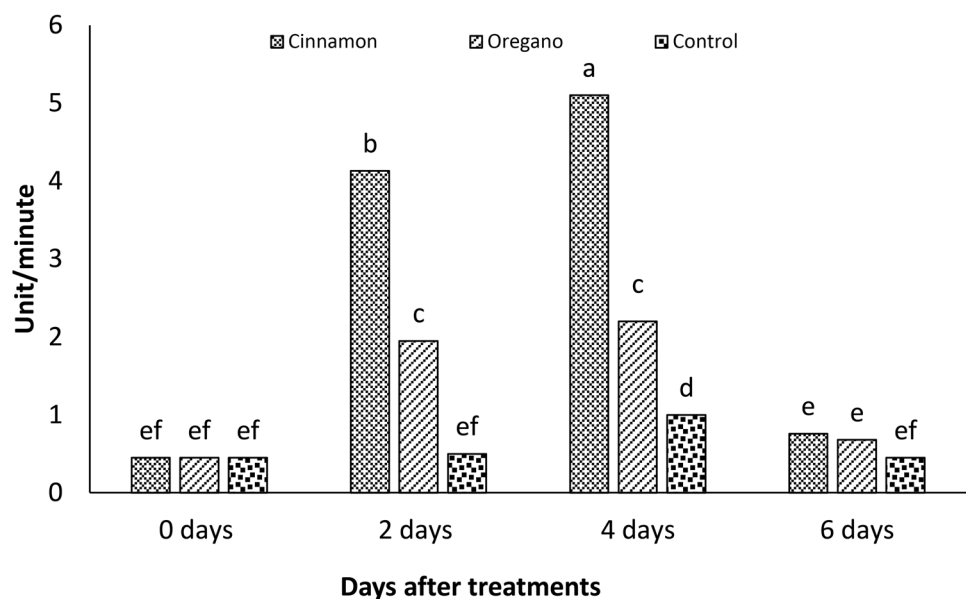
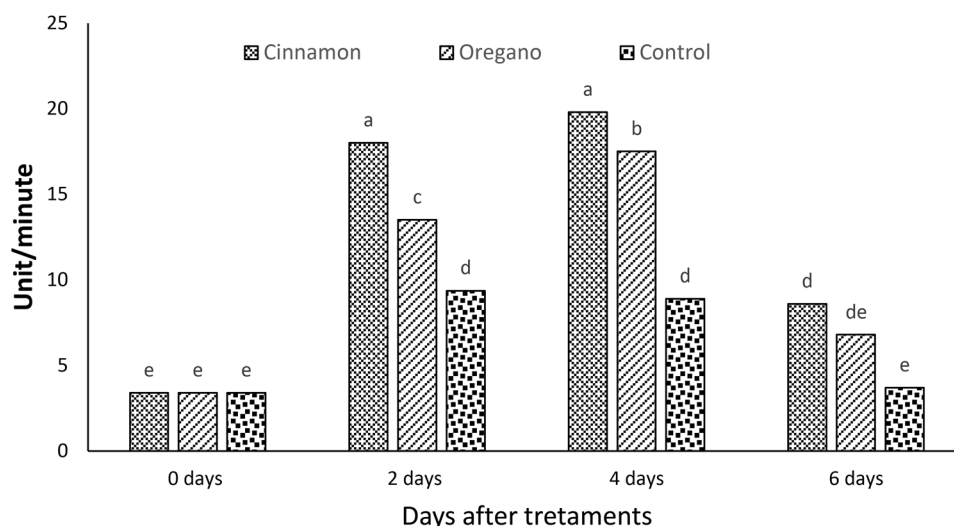


Fig. 3 Effect of treatments with essential oils of cinnamon (*Cinnamomum verum* L.) and oregano (*Origanum vulgare* L.) on polyphenoloxidase activity (U/min/g fw) in extracts of “Taify” grape tissues. Values in the column followed by different letters indicate significant differences among treatments according to a least significant differences test ($P=0.05$)



Effects of cinnamon and oregano EOs on chemical properties of fruits

Total soluble solids content

The data in Fig. 6 show that treating infected “Taify” grape fruits with cinnamon and oregano oil at 800 $\mu\text{L/L}$ did not increase TSS compared with the control. After four days, TSS steadily declined until the sixth day. At the end of this time, the highest TSS levels were found in untreated fruits, while the lowest levels were seen in treated fruits.

Vitamin C content

Figure 7 shows that in case of infected “Taify” grape fruits treated with cinnamon and oregano oil at 800 $\mu\text{L/L}$, there was no significant difference in vitamin C content (VC) between the treatments and controls. At day 4, the vitamin C concentration started to decline until day 6. Nevertheless, fruits sprayed with EOs showed higher levels of VC than the control. It was apparent that each oil treatment showed the same increment in VC.

Fig. 4 Effect of treatments with essential oils of cinnamon (*Cinnamomum verum* L.) and oregano (*Origanum vulgare* L.) on phenol content (phenols [mg/g fw] in extracts of “Taify” grape tissues. Values in the column followed by different letters indicate significant differences among treatments according to a least significant differences test ($P=0.05$)

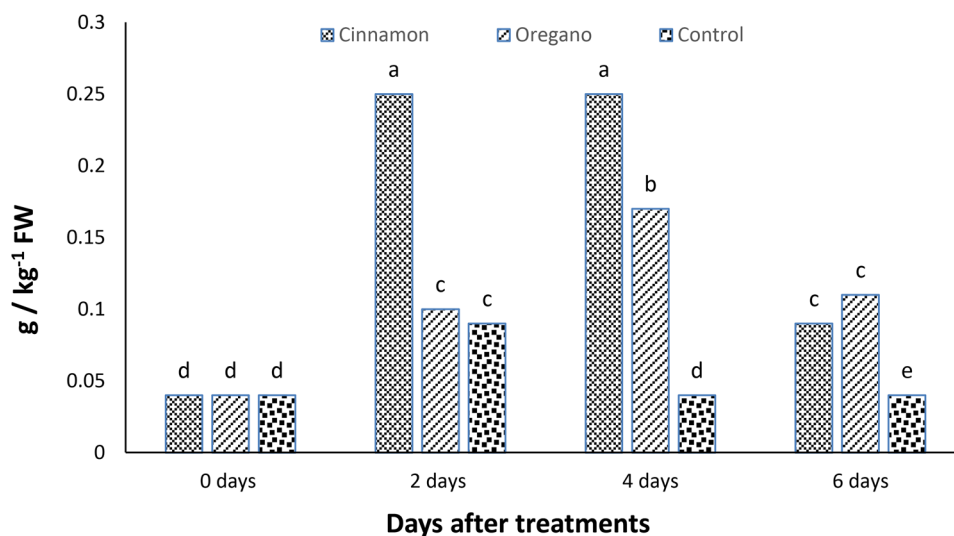
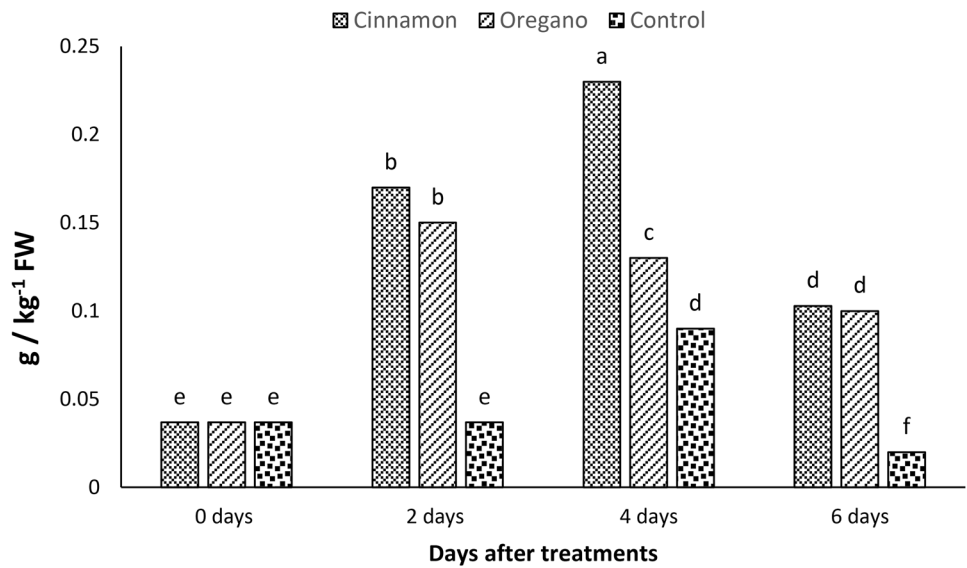


Fig. 5 Effect of treatments with essential oils of cinnamon (*Cinnamomum verum* L.) and oregano (*Origanum vulgare* L.) on flavonoids (mg/g fw) in extracts of “Taify” grape tissues. Values in the column followed by different letters indicate significant differences among treatments according to a least significant differences test ($P=0.05$)



Titrateable acidity (TS)

At the beginning of the storage period, the amount of titrateable acidity (TA) in table grape “Taify” did not change in

fruits treated with cinnamon or oregano oils or untreated fruits (Fig. 8). With the passage of time, the quantity of TA in table grapes dropped. However, the TA increased in the coated fruits compared with the untreated fruits.

Fig. 6 Effects of treatment with essential oils of cinnamon (*Cinnamomum verum* L.) and oregano (*Origanum vulgare* L.) on total soluble solids (%) in extracts of “Taify” grape tissues. Values in the column followed by different letters indicate significant differences among treatments according to a least significant differences test ($P=0.05$)

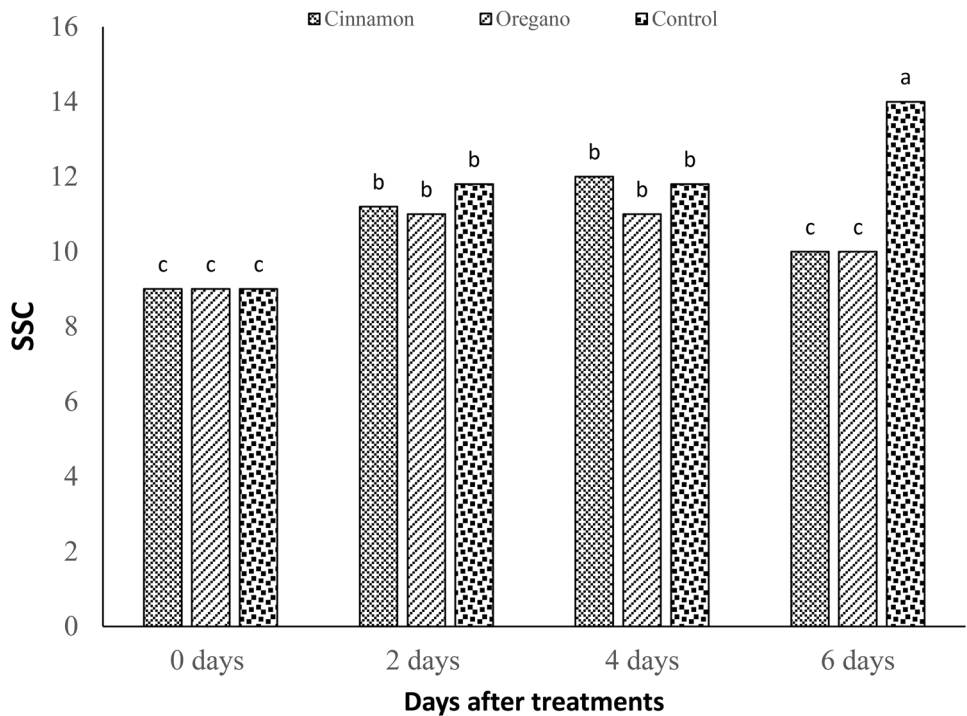
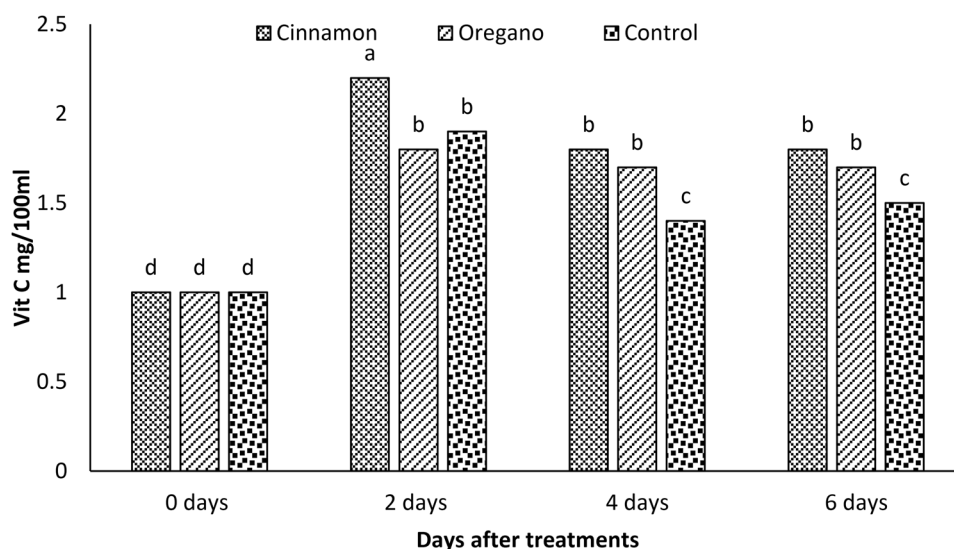


Fig. 7 Effect of treatments with essential oils of cinnamon (*Cinnamomum verum* L.) and oregano (*Origanum vulgare* L.) on Vitamin C (mg/100 mL) of “Taify” grape tissues. Values in the column followed by different letters indicate significant differences among treatments according to a least significant differences test ($P=0.05$)



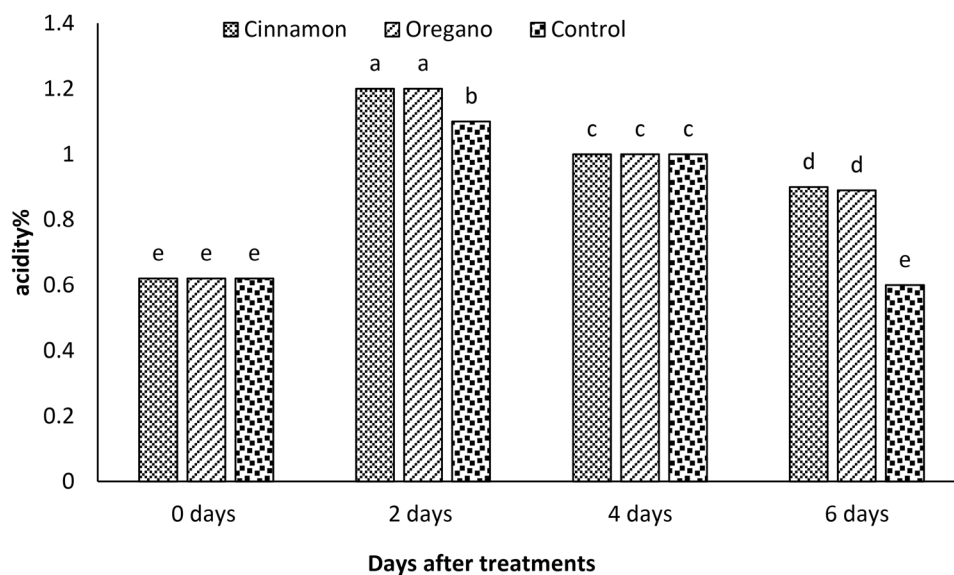
Discussion

In recent years, there has been a lot of interest in finding reasonably safe bio-fungicides, such as EOs, to manage plant diseases in agriculture. In our study, we used various concentrations of cinnamon and oregano EOs (100, 200, 400, 600, 800, and 1000 $\mu\text{L/L}$) against *B. cinerea*, which causes gray mold disease of grape fruits. The most effective concentrations of both EOs for inhibition of *B. cinerea* growth were 800 and 1000 $\mu\text{L/L}$. Our results are accordance with those of Vitoratos et al. (2013), who noted that in the presence of lemon and oregano EOs, *B. cinerea* did not display any mycelial growth. Increased doses of EOs (*Aloysia citriodora*, *Cymbopogon winterianus*, and *Ocimum americanum*) made them fungitoxic and reduced

the mycelial growth of *Colletotrichum* sp., *B. cinerea*, and *Monilinia fructicola* (Fontana et al. 2021). Pansera et al. (2015) noted that a higher dose of *Cymbopogon camphora* and *Eucalyptus globulus* EOs was more effective at reducing mycelial growth of *M. fructicola* compared to lower doses.

Cinnamon and oregano oils decreased spore germination by approximately 65.2% in the present study. EOs generated from aromatic and medicinal plants have a wide range of antibacterial activities (Perdonés et al. 2012; Khalili et al. 2015; Campos-Requena et al. 2017), and oil content, structure, and functional groups all play a role in antibacterial and antifungal activities (Feng and Zheng 2007). Most antifungal components have been shown to inhibit conidial germination, resulting in the fungus being killed (Tzortzakis

Fig. 8 Effect of treatments with essential oils of cinnamon (*Cinnamomum verum* L.) and oregano (*Origanum vulgare* L.) on titratable acidity (TA%) of “Taify” grape tissues. Values in the column followed by different letters indicate significant differences among treatments according to a least significant differences test ($P=0.05$)



2010). Zamani-Zadeh et al. (2014) mentioned that essential oil vapor treatments that suppress mycelial growth could contribute to preventing pathogen spread by reducing the spore density on the surface.

Our *in vivo* study showed that grape fruits treated with cinnamon or oregano oil at 800 $\mu\text{L/L}$ could significantly reduce the percentage of infected grape fruits compared to the control. Oregano oil suppressed the disease severity of gray mold more effectively than cinnamon oil. Our results are in line with Vitoratos et al. (2013), who reported that oregano EOs are able to inhibit gray mold of tomato caused by *B. cinerea*. Previous studies have found that essential oil compounds, such as thymol, carvacrol, eugenol and menthol, can extend the shelf life of table grapes (Martinez-Romero et al. 2007; Kavooosi et al. 2014). The immediate mechanism of EOs is linked to the lipophilic property of oil molecules, which firmly attach to membranes, thereby altering selectivity and aiding penetration through membranes, which results in microbial cell energy loss (Knaak and Fiuza 2010). Essential oils suppress fungal growth, often by preventing hyphal growth and sporulation, interrupting nutritional absorption and metabolism, disturbing the plasma membrane, damaging the mitochondrial structure, and interfering with enzymatic and respiratory processes (Patel and Jasrai 2011).

Treating “Taify” grape fruits with each essential oil at 800 $\mu\text{L/L}$ increased the activity of the POD and PPO at two days after treatment compared to the untreated control. At four days, enzyme activity increased, and at six days, it decreased. The cinnamon oil showed higher PPO activity than oregano oil. The findings of this study are in agreement with those of Chen et al. (2019), who found that treatment of fruit with clove essential oil fruit increased the activity of PAL, POD, and PPO compared to the control. Higher levels of antioxidants, peroxidase (POD), and polyphenoloxidase are seen in lemon fruits treated with both *Schwanniomyces vanrijiae* and ethanolic preparations of propolis than in untreated controls (Abo-Elyousr et al. 2021). Plant resistance to various stresses is frequently demonstrated by an increase in defensive enzymes such as POD (Youssef et al. 2020; Abo-Elyousr et al. 2021). Generally, activation of the PR proteins PAL, POD, and PPO, which are involved in the biosynthesis and oxidation of phenolic compounds, is associated with plant disease resistance (Livak and Schmittgen 2001; Sallam et al. 2021). PPO plays a very important role in activating oxygen metabolism and oxidizes phenolic compounds in plants to hazardous quinones to prevent pathogen growth, while PAL and POD are critical enzymes in the phenylpropanoid metabolic pathway that promote synthesis (Shao et al. 2013). Banani et al. (2018) found that apple fruits treated with essential oil induced resistance against *B. cinerea* through the priming of defense responses in apple fruits.

In the present study, EOs increased the total phenol content and total flavonoid content in grape cv. “Taify” fruits at 2 to 4 days after treatment as compared to control fruits. The total phenol content and total flavonoid decreased at 6 days. Treating fruits with cinnamon oil resulted in higher total phenol content compared to oregano oil. There was no major difference in total flavonoid content between cinnamon and oregano oil. The results are in agreement with those of Abo-Elyousr et al. (2021), who showed that lemons treated with both *S. vanrijiae* and ethanolic preparations of propolis had the highest total phenol and total flavonoid levels. By modifying cellular processes, phenols and flavonoids preserve many cellular components from destruction and play a crucial role in plant growth and development. Additionally, increased enzymatic activity can result in increased phenolic content and antioxidant enzyme activity (Chuying et al. 2019). POD activity helps to control the production of hydrogen peroxide in the cell wall, which is essential for phenolic group cross-linking in response to external stresses (Raimbault et al. 2011; Youssef et al. 2020).

The same amount of TSS in table grape fruits treated with cinnamon and oregano oil and the control was observed. TSS steadily decreased in all treatments, but the highest TSS levels were found in uncoated fruits, while the lowest were seen in treated fruits. The capacity of the coating material to minimize the migration of water from the fruit surface to the external area was associated with a reduction in TSS levels of coated fruits (Sallam et al. 2012; Aloui and Khwaldia 2016).

Vitamin C levels were similar in treated and untreated fruits, but decreased as storage time increased. Salimi et al. (2013) reported that application of basil and wild mint oil the increased content of vitamin C compared to untreated fruits. The oxidation of superoxide and hydroxyl radicals in strawberry fruits may be responsible for this reduction (Martínez et al. 2018; Zhang et al. 2018). The amounts of ascorbic acid in fruits treated with different EOs were higher at the end of the storage period compared to untreated fruits. Vitamin C concentration is crucial for fruit preservation during storage (Zhang et al. 2018). Our results are in line with those of Bagy et al. (2021), who suggested that treating orange fruits with EOs could effectively prevent vitamin C (ascorbic acid) loss during storage. This could be due to the antioxidant capabilities of essential oil coatings, which decrease oxygen diffusion, reduce respiration rate (Shehata et al. 2020), and thus reduce ascorbic acid oxidation (Aloui and Khwaldia 2016; Marín et al. 2016).

In the present study, no differences in TA were observed between fruits treated with EOs and untreated fruits. The sprayed fruits had a higher level of TA at the end of the storage period than the untreated fruits. Edible films containing EOs decrease the loss of water, respiration, and microbial

growth, resulting in lower organic acid consumption (i.e., titratable acid) in the respiratory metabolic processes of strawberry fruits. This effectively minimizes titratable acid loss, extending the shelf life of stored fruit after harvest (Gol et al. 2013; Badawy et al. 2011; Shehata et al. 2020).

Conclusions

These results demonstrate the *in vitro* and the *in vivo* antifungal activities of oregano and cinnamon oils against *B. cinerea* and their potential use as biological fungicides for the control of postharvest fungal rot disease caused by *B. cinerea* on grape fruits and may be applied as a future ecofriendly alternative to synthetic fungicides for controlling gray mold disease.

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Authors' contributions All authors contributed equally in the manuscript. NMA suggested the idea of the work and contributed to data curation and their validation as well as writing original draft. ADQ contributed to the formal analysis of the data, M.E.I and KAMA contributed to the reviewing and editing the manuscript. All authors reviewed and approved the final version of the manuscript.

Declarations

Ethical responsibility Our manuscript is original research, and it is not submitted to full or in parts to other journal for publication.

Informed consent All authors have reviewed the manuscript and approved the final version of manuscript before submission.

Conflict of interest The authors declare that they do not have any actual or potential conflict of interest.

Human and animal studies The research did not involve any studies with human participants or animal as experimental model.

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