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Inhibitory Effect of Some Plant Essential Oils against Corn Stalk Rot and Ear Rot

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Abstract: Yield losses are experienced in corn fields due to stalk and ear rot disease. The fungicides used against the disease can not show the desired effect and the problem of resistance arises over time. For this reason, it is necessary to develop new strategies in the control against the disease. In this study, it was aim to determine the inhibitory effects of essential oils of rosemary, black cumin, cumin and sandalwood, and ginger against *Fusarium verticillioides* and *Fusarium pseudograminearum* under *in vitro* conditions. The essential oils were applied in doses of 0.25, 0.5, 1, 2 and 4 µl mL^{-1}. PDA medium free of essential oils was used as negative control and PDA medium containing commercial fungicide (80 g l^{-1} Triticonazole, 40 g l^{-1} Pyraclostrobin-BASF company) was used as positive control. The experiment was carried out in a randomized plot design with three replications. In addition, chemical content analysis of essential oils was determined by GC-MS method. As a result of the study, as the dose of rosemary, black cumin, sandalwood and ginger essential oils increased, their inhibitory effects against both pathogens increased, and these effects were found close to each other. While the highest inhibitory effect against *F. verticillioides* and *F. pseudograminearum* was detected in the positive control treatment of cumin essential oil, 82.1% and 78.9%, respectively, this effect was found to be 74.6% and 68.3% in 4 µl mL^{-1} dose, respectively. However, more detailed studies should be carried out with these essential oils under field conditions.

Keywords: Corn, fungal colony, *Fusarium pseudograminearum*, *Fusarium verticillioides*, plant essential oils

Bazı Bitkisel Uçucu Yağların Mısır Sap ve Koçan Çürüklüğüne Karşı Engelleyici Etkisi

Öz: Mısır ekim alanlarında sap ve koçan çürüklüğü hastalığı sebebiyle verim kayıpları yaşanmaktadır. Hastalığa karşı kullanılan fungisitler istenilen etkiyi gösterememekte ve zamanla dayanıklılık sorunu ortaya çıkmaktadır. Bu sebeple hastalığa karşı mücadelede yeni stratejilerin geliştirilmesi gerekmektedir. ,Çalışmada *in vitro* koşullarda biberiye, çörekotu, kimyon, sandal ağacı ve zencefi uçucu yağlarının mısırda *Fusarium verticillioides* ve *Fusarium pseudograminearum*'a karşı engelleyici etkisinin belirlenmesi amaçlanmıştır. Uçucu yağlar 0.25, 0.5, 1, 2 ve 4 µl mL^{-1}'lik dozlarda uygulanmıştır. Uçucu yağlardan ari PDA besiyeri negatif kontrol, ticari fungisit (80 g l^{-1} Triticonazole, 40 g l^{-1} Pyraclostrobin-BASF company) içeren PDA besiyeri ise pozitif kontrol olarak kullanılmıştır. Deneme, tesadüf parselleri deneme deseninde 3 tekerrürlü olarak yürütülmüştür. Ayrıca uçucu yağların kimyasal içerik analizi GC-MS metodu ile belirlenmiştir. Çalışma sonucunda, biberiye, çörek otu, sandal ağacı ve zencefil uçucu yağlarının dozu arttıkça her iki patojene karşı engelleyici etkileri artmış ve bu etkiler birbirlerine yakın bulunmuştur. *F. verticillioides* ve *F. pseudograminearum*'a karşı en yüksek engelleyici etki kimyon uçucu yağının pozitif kontrol uygulamasında sırasıyla %82.1 ve %78.9 oranında saptanırken, bu etki 4 µl mL^{-1} dozunda sırasıyla %74.6 ve %68.3 olarak saptanmıştır. Bununla birlikte, bu uçucu yağlar ile tarla şartlarında daha detaylı çalışmaların yürütülmesi gerekmektedir.

Anahtar Kelimeler: Bitki uçucu yağları, fungal koloni, *Fusarium pseudograminearum*, *Fusarium verticillioides*, mısır

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1. Introduction

Corn, which is included in the Maydeae tribe of the Gramineae family, is a plant that shows the highest yield in the world among all cool climate and hot climate cereals, can use solar energy best (C4 plant) and can produce the most dry matter per unit area (Kırtok, 1998). The corn plant, which has a wide production, has left many industrial plants behind in terms of usage purposes. Maize is mostly grown as fodder, silage or grain to feed livestock (Klopfenstein et al., 2013). Even while animal feed accounts for the majority of the world's maize output, it is also a staple of human diet, particularly in the form of corn flour, corn bread, and corn syrup (Shah et al., 2016). Corn is also used in the plastic and fabric industry, in the production of biofuels, as a nutrient medium to grow many types of microorganisms, and as a food dye (Raut et al., 2015; Someshwar et al., 2018; Aly Emam et al., 2020). According to world data, approximately 1 billion tons of corn is produced, and the United States is the world's largest corn producer with an annual production of approximately 380 billion tons. Corn is cultivated on 692 thousand hectares of land and 6.5 million tons of corn are produced in Turkey, which comes in at number 19 in terms of cultivation area and production (TSI, 2021).

In corn farming, serious yield losses are experienced every year due to biotic and abiotic stress factors. Fungal, viral and bacterial disease factors from biotic stress factors cause significant losses in corn yield (Peng et al., 2021). Many *Fusarium* species cause significant yield losses in root, root collar, stem, leaf or cob of cool climate cereals (barley, wheat, oats and rye etc.) and warm climate cereals (corn, paddy and sorghum etc.) (Desmond et al., 2008). These diseases are seen in grain production regions in the world and cause 10-30% yield loss in Europe every year (Foroud et al., 2014). According to reports, the most significant diseases that reduce maize yields worldwide in corn-producing regions are *Fusarium* stem and ear rots. (Miller, 1994; Bottalico, 1998; Edwards, 2004). Bends and twists around the stem axis are typically seen in plants due to stem and ear rot disease. After the fungus enters the mature plant roots, it develops in the bark tissue, goes to the trunk and can cause death in the seedling stage. *Fusarium verticilloides* (Sacc) Nirenberg (Syn: *F. moniliforme*) forms whitish-pink micelles on corncobs (Tiru et al., 2021). Without causing any symptoms, the pathogen that was present in the seed can be discovered endophytically in the plant (Burgess and Wayn, 2012). Pathogenic fungi produce toxins at levels harmful to human and animal health (Gurdaswani and Ghag, 2020). Wrinkled white heads may develop in grains infected with *Fusarium pseudograminearum*, and as a result of an effect on grain development, grain yield, grain number, grain weight and stalk weight decrease (Chekali et al., 2016). It is reported that *F. verticilloides* produces Zearalenone (ZEA), Fumonisin (FUM) and Deoxynivenol (DON) toxins in maize. (Blandino et al., 2009; Miadener et al., 2010; Mukanga et al., 2010; Goertz et al., 2010). The amount of toxin formed according to the severity of the disease harms human and animal health (Munkvold, 2001). High doses of ZEA cause persistent oestus and infertility, especially in animals (Fink-Gremmels, 1999).

Fungicides containing various active ingredients (pyraclostrobin, sedaxana and triticonazole) are used in the chemical control against stalk and ear rot in corn (Anonymous, 2021). Despite the fact that synthetic fungicides are effective, they have made it necessary to develop non-chemical control methods in the control against disease factors because they over time cause the emergence of resistant strains in different pathogens, leading to epidemics and environmental pollution by upsetting the natural balance (Temur and Tiryaki, 2012). Essential oils are complex mixtures and contain a number of chemical metabolites with potential antifungal properties (Nazzaro et al., 2017). Antifungal effects are attributed to the predominant components in their chemical structures. Monoterpenes, diterpenes, sesquiterpenes and their oxygen derivatives in hydrocarbon structure, as well as alcohols, aldehydes, esters, ketones, phenolic and oxidized components can also be found in the structures of essential oils (İşcan, 2002). The antifungal effects of *Rosmarinus officinalis* L., *Nigella sativa* L., *Cuminum cyminum* L., and *Zingiber officinale* essential oils against various pathogens have been determined in studies (Ozcan et al., 2008; Mahmoudvand et al., 2014; Basım and Basım, 2017; Ghasemi et al., 2018; Castro et al., 2020). In studies conducted to determine the antifungal effect against *F. verticillioides,* the antifungal effect of essential oils of *R. officinalis, C. cyminum* and *Z. officinale* has been reported (da Silva Bomfim et al., 2015; Khosravi et al., 2015; Castro et al., 2020). In the literature review, no studies were found to defined the antifungal effect of essential oils of *A. andrachne, R. officinalis, C. cyminum, N. sativa* and *Z. officinale* against *F. pseudograminearum*, and of *A. andrachne* and *N. sativa* against *F. verticillioides*.

In this study, it was aimed to determine the inhibitory effect of different concentrations of five essential oils (rosemary, black cumin, cumin, sandalwood and ginger) against *F. verticillioides* and *F. pseudograminearum* in maize under *in vitro* conditions.

2. Materials and Methods

2.1. Fungal isolates

In the experiment, *F. verticillioides* and *F. pseudograminearum* isolates with known virulence and isolated from maize plant were obtained from the fungal culture collection of the General Directorate of Agricultural Research and Policies, Directorate of Plant

Protection Central Research Institute. Fungal isolates were grown on potato dextrose agar medium (PDA-Difco) in a cooled incubator (25±1 °C temperature; 12 hours of darkness/12 hours of light) and stored at +4 °C in the refrigerator.

2.2. Plant materials

In the study, essential oils of plants belonging to different families such as rosemary (*R. officinalis* L), black cumin (*N. sativa* L), cumin (*C. cyminum* L.), sandalwood (*A. andrachne* L.) and ginger (*Z. officinale*) were used. It was produced by a private company by steam distillation method and used as plant material (Table 1). The essential oils were stored in the refrigerator at +4 °C in dark colored and tightly closed bottles until used.

2.3. Determination of essential oils by gas chromatography/mass spectrometry (GC-MS)

GC-MS analysis of essential oil components was carried out at Süleyman Demirel University Innovative Technologies Application and Research Center (YETEM). Component analyzes were performed on a gas chromatography-mass spectrometry (GC-MS) (Shimadzu 2010 SE, Kyoto Japan) instrument. The individual peaks were identified by comparing linear retention indices (LRI) as well as comparing mass spectra with the Wiley library (Wiley, New York, NY, USA) and the NIST mass spectral database (Gaithersburg, MD, USA) (Semiz et al., 2016).

2.4. Inhibitory effect of essential oils on fungal growth *in vitro* conditions

The inhibitory effect of rosemary, black cumin, cumin, sandalwood and ginger essential oils belonging to different families against *F. verticillioides* and *F. pseudograminearum* was investigated according to the contact application test with *in vitro* petri dishes. Essential oils of rosemary, black cumin, cumin, sandalwood and ginger plants were dissolved in ethanol solution at a 1:2 ratio and poured into sterile petri dishes (10 cm diameter) with PDA at different concentrations, instead of PDA medium sterilized at 121 °C for 15 minutes in autoclaved and cooled (Table 2). The 7-day-old fungal isolates that had previously been cultivated in PDA medium were cut into discs with a 5 mm diameter fungus discs and put in the middle of petri dishes with essential oil + PDA medium. PDA medium free from essential oils was used as negative control, and licensed fungicide (80 g l^{-1} Triticonazole, 40 g l^{-1} Pyraclostrobin-BASF company) against stalk and ear rot disease in maize was used at recommended dose as positive control. The petri dishes were incubated at 25±1 °C for 10 days after inoculation and then covered with parafilm. The measurement of the colony diameter was made by measuring the diameter of the fungus colony in separate directions perpendicular to each other (Benjilali et al., 1984). The inhibitory percentage of different doses of essential oil was calculated using the formula MGI (%) = [(C-T) /C] x 100, where MGI: Mycelial growth inhibitory percentage, C and T, respectively, are the diameter of colonies in control petri dish (mm) and the diameter of colonies treatment petri dish (mm) (Dev et al., 2004). The experiment was carried out in a randomized plot design with 3 replications.

2.5. Statistical methods

Statistical analyses of the data were performed with the JMP IN packet statistic program (SAS Institute, Carry, NC, 13.0 PC version). Analysis of variance (one-way ANOVA) was carried out to determine the effects of the treatments. When the treatment effects were statistically significantly ($p \leq 0.01$), the Duncan's multiple range test was used for means separations.

3. Results and Discussion

The GC-MS analysis results were examined in the study, a total of 80 active substances were determined in 5 different plant essential oils (Table 3). Eucalyptol (1,8-Cineole) was the highest in *N. sativa* essential oil component with a rate of 48.28%, followed by cuminaldehyde (31.44%) in *C. cyminum* (Table 3).

Table 1. Common name, scientific name, family, brand name and company name of the plant materials used in this study

Common name	Scientific name	Family	Brand name	Company name
Rosemary	*Rosmarinus officinalis* L	Lamiaceae	Rosemary Oil	
Black cumin	*Nigella sativa* L.	Ranunculaceae	Black Cumin Oil	Arpaş Arifoğlu Marketing,
Cumin	*Cuminum cyminum* L.	Apiaceae	Cumin Oil	Distribution and Trade Inc.
Sandalwood	*Arbutus andrachne* L.	Ericaeeae	Sandalwood Oil	Avcılar-İstanbul/TURKEY
Ginger	*Zingiber officinale*	Zingiberaceae	Ginger Oil	

Table 2. Treatment of plant essential oils against *F. verticillioides* and *F. pseudograminearum*

	Application	Doses (µl mL^{-1})
	Rosemary (*Rosmarinus officinalis* L)	0.25
	Black cumin (*Nigella sativa* L.)	0.5
Essential oils	Cumin (*Cuminum cyminum* L.)	1
	Sandalwood (*Arbutus andrachne* L.)	2
	Ginger (*Zingiber officinale*)	4

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Table 3. GC-MS analysis results of essential oils of rosemary (*R. officinalis*), black cumin (*N. sativa*), cumin (*C. cyminum*), sandalwood (*A. andrachne*) and ginger (*Z. officinale*)

			R. officinalis	*N. sativa* [c]	*C. cyminum* [c]	*A. andrachne*	*Z. officinale* [c]
No	**Compound name** [a]	**LRI** [b]	**% of the essential oil**				
1	Tricyclene	924	1.05	0.77	0.04	0.21	1.44
2	alpha- Thujene	927	0.23	2.40	0.32	0.27	0.16
3	alpha - Pinene	933	20.06	14.78	1.68	4.90	7.01
4	beta- Fenchene	942	0.10	-	-	-	0.12
5	Camphene	953	5.02	4.77	0.24	1.62	12.33
6	Sabinene	972	1.15	2.23	0.32	0.61	0.51
7	beta- Pinene	978	2.96	9.07	16.03	2.96	2.11
8	4-Methyl-1-hepten-5-one	986	-	-	-	-	0.54
9	beta- Myrcene	991	0.48	-	0.69	-	1.55
10	Octanal	1006	-	-	-	-	0.14
11	Phellandrene	1007	0.16	-	0.44	-	0.39
12	DELTA.3-Carene	1009	0.45	-	0.05	-	0.04
13	alpha- Terpinene	1018	0.72	-	0.18	-	0.13
14	Cymol	1025	4.68	8.41	14.73	1.85	1.17
15	Limonene	1030	8.66	3.10	1.19	1.36	12.72
16	Eucalyptol (1,8-Cineole)	1052	24.02	48.28	1.93	15.10	16.27
17	gamma-Terpinene	1058	2.10	3.36	17.79	1.41	0.70
18	trans-Sabinene hydrate	1088	-	0.52	0.03	0.26	0.20
19	alpha- Terpinolen	1096	0.35	-	0.13	-	0.28
20	Dimethylstyrene (alpha-para)	1104	-	-	0.52	-	0.65
21	Linalool	1114	1.94	-	-	-	-
22	Chrysanthenone	1133	-	-	-	0.34	-
23	Carveol	1152	-	-	0.10	-	-
24	Camphor	1157	2.01	2.32	0.09	0.93	0.87
25	Isoborneol	1165	15.84	-	-	-	-
26	4-Terpineol	1193	0.49	-	0.28	-	0.18
27	Dimethylbenzylcarbinyl acetate (DMBCA)	1200	-	-	0.35	-	0.62
28	alpha- Terpineol	1207	4.31	-	-	-	-
29	Perilla alcohol	1208	-	-	0.85	-	-
30	Dihydrocarvone	1210	-	-	0.11	-	-
31	alpha- Terpinyl acetate	1213	0.60	-	-	-	-
32	Z-Citral	1238	-	-	-	-	1.88
33	Cuminaldehyde	1247	-	-	31.44	-	-
34	Carvotanacetone	1260	-	-	0.33	-	-
35	E-Citral	1268	-	-	-	-	2.25
36	Phellandral	1277	-	-	0.34	-	-
37	Bornyl acetate	1287	2.00	-	-	-	-
38	2-Undecanone	1294	-	-	-	-	0.17
39	2-Caren-10-al	1298	-	-	6.84	-	-
40	1-Phenylpropane-1,3-diol	1302	-	-	0.89	-	-
41	Thymol	1307	-	-	0.10	-	-
42	Carvacrol	1317	-	-	0.05	-	-
43	R(+)-Limonen	1358	-	-	-	2.93	-
44	Citronellyl acetate	1363	-	-	-	-	0.37
45	alpha- Copaene	1375	-	-	-	-	0.25
46	Hydrocoumarin	1386	-	-	-	5.93	-
47	gamma- Cadinene	1388	-	-	0.09	-	-
48	Linalyl acetate	1392	-	-	-	-	0.93
49	beta- Elemene	1400	-	-	-	-	0.33
50	alpha- Zingiberene	1414	-	-	-	-	0.09
51	Caryophyllene	1428	0.63	-	0.12	-	-
52	Coumarin	1438	-	-	-	5.94	-
53	Germacrene B	1439	-	-	-	-	0.15
54	Farnesene ((E)-, beta)	1466	-	-	0.05	-	-
55	alpha- Cedrene	1483	-	-	1.32	-	-
56	Germacrene D	1490	-	-	-	-	0.45
57	Curcumene	1491	-	-	-	-	4.30
58	Alloaromadendrene	1503	-	-	-	-	0.52
59	Sesquithujene (7-epi)	1506	-	-	-	-	17.72
60	alpha- Farnesene	1517	-	-	-	-	1.40
61	beta- Bisabolene	1519	-	-	-	-	3.80
62	beta- Sesquiphellandrene	1534	-	-	-	-	3.80
63	Ethyl phthalate	1592	-	-	-	7.23	-
64	Carotol	1601	-	-	0.05	-	-
65	Cedryl methyl ether	1624	-	-	-	4.56	-
66	Widdrene	1717	-	-	-	2.30	-
67	alpha.-Hexylcinnamaldehyde	1754	-	-	-	1.35	-
68	Cedrene	1768	-	-	-	18.63	-
69	Cedryl acetate	1771	-	-	-	2.48	-
70	beta- Guaien	1777	-	-	-	0.85	-
71	Aromadendrene	1790	-	-	-	0.39	-

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72	Hexamethyl-pyranoindane	1847	-	-	-	6.99	-
73	Benzene, 1-(1,1-dimethylethyl)-3,5-dimethyl-2,4,6-trinitro-	1852	-	-	-	1.11	-
74	Tetralin (6-Acetyl-, 1,1,2,4,4,7-hexamethyl-)	1855	-	-	-	4.00	-
75	Musk ketone	1961	-	-	-	3.48	-
76	Tetracosane	2400	-	-	0.07	-	-
77	Pentacosane	2500	-	-	0.08	-	-
78	Hexacosane	2600	-	-	0.07	-	-
79	Heptacosane	2700	-	-	0.06	-	-
80	Nonacosane	2900	-	-	0.02	-	-
			100	100	100	100	100

[a] Compounds listed in order their elution, [b] LRI: Linear retention index, [c] GC-MS analysis results are shared in the article of Sağlan et al. (2022).

The effects of five various concentrations (0.25, 0.5, 1, 2 and 4 µl mL^{-1}) of rosemary, black cumin, cumin, sandalwood, and ginger plant essential oils on *F. verticillioides* and *F. pseudograminearum* on mycelial growth and inhibition rates are shown in Table 4 for the study conducted *in vitro* conditions. In comparison to the negative control, it was discovered that the inhibitory effects of rosemary, black cumin, cumin, sandalwood, and ginger essential oils on the mycelial growth of *F. verticillioides* and *F. pseudograminearum* were statistically significant (P≤0.01). Depending on the dose increase, each essential oil utilized in the study reduced the pathogens mycelial development at a different rate. Positive control treatment of rosemary essential oil showed 81.7% and 80.4% effects on tested *F. verticillioides* and *F. pseudograminearum*, respectively, while high dose (4 µl mL^{-1}) treatment followed the next highest effect (64.8% and 60.6%). In other doses of rosemary essential oil, inhibition of mycelial growth of *F. verticillioides* was between 19.3% and 50.9%, while inhibition of mycelial growth of *F. pseudograminearum* was between 15.7% and 47.4%. While the highest effect on *F. verticillioides* and *F. pseudograminearum* in black cumin essential oil was detected in the positive control treatment at 80.4% and 73.0%, respectively, this treatment was followed by 4 µl mL^{-1} treatment (72.5% and 62.7%). While other doses of black cumin essential oil inhibited mycelial growth of *F. verticillioides* between 19.8% and 53.7%, it inhibited mycelial growth of *F. pseudograminearum* between 15.0% and 50.8%. Positive control application of cumin essential oil showed 82.1% and 78.9% effects against *F. verticillioides* and *F. pseudograminearum*, respectively, while this effect was 74.6% and 68.3%, respectively, in 4 µl mL^{-1} treatments. While other doses of cumin essential oil inhibited mycelial growth of *F. verticillioides* between 14.5% and 57.1%, it inhibited mycelial growth of *F. pseudograminearum* between 13.9% and 51.6%. Positive control treatment of sandalwood essential oil showed the highest inhibitory effect against *F. verticillioides* (61.9%), while this effect was found to be 58.0% in *F. pseudograminearum*. In five doses of sandalwood essential oil, inhibition of mycelial growth of *F. verticillioides* was between 16.3% and 51.5%, while inhibition of mycelial growth of *F. pseudograminearum* was between 14.5% and 48.9%. The highest inhibitory effect in ginger essential oil was determined against *F. verticillioides* and *F. pseudograminearum* in the positive control treatment at a rate of 79.6% and 74.7%, respectively. Similar antifungal effects were seen against both pathogens in all five doses of ginger essential oil. While the inhibition of mycelial growth of *F. verticillioides* was between 20.5% and 66.2%, inhibition of mycelial growth of *F. pseudograminearum* was between 18.3% and 61.6% (Table 4).

In the study, cumin essential oil was determined as the most effective essential oil inhibiting mycelial growth of *F. verticillioides* and *F. pseudograminearum*, compared to rosemary, black cumin, sandalwood and ginger essential oils. Positive control and high dose (4 µl mL^{-1}) application of cumin essential oil showed a high inhibitory effect against *F. verticillioides* and *F. pseudograminearum*, and the antifungal effects of the other four essential oils were found to be close to each other, depending on the pathogen and dose.

The main components of the essential oil and extract from rosemary differ. According to the results of GC-MS analysis, the main components of rosemary essential oil were determined as eucalyptol (1,8-cineole) (24.02%), α-pinene (20.06%) and isoborneol (15.84%) (Table 3). The effects of essential oils depend on their chemical composition, which is influenced by many factors such as plant genotype, geographic region, environmental factors and agronomic conditions (Yeşil Çeliktaş et al., 2007). In this context, the antimicrobial effects of essential oils; it determines the composition, structure and functional groups of these compounds (Omidbeygi et al., 2007). Rosemary (*R. officinalis* L.) from the Laminacae family is an important medicinal and aromatic plant species. Rosemary has biological effects such as antifungal, antimicrobial, antioxidant and antiviral. In a conducted study, it was reported that *R. officinalis* significantly reduced mycelial growth of *F. verticillioides* (da Silva Bomfim et al., 2015). Another study also showed that *R. officinalis* inhibited the growth and conidial production of *F. verticillioides* (Achimón et al., 2021). Black cumin, one of the most useful essential oils known, is a perennial herbaceous plant used in the food, medicine and cosmetics industries. In the content of *N. sativa* essential oil, 12 components were found together with eucalyptol (1,8-cineole) (48.28%) and α-pinene (14.78%), which are high in content. It has been reported that eucalyptol, the main component of *N. sativa* essential oil, completely inhibits the growth of various fungal species (Naz, 2011).

In addition, essential oil of black cumin seed completely inhibits mycelial growth of *F. verticillioides* (Elgorban et al., 2015). Cumin with important medicinal properties is a well-known aromatic plant. GC-MS results showed that cuminaldehyde (31.44%), γ-terpipene (17.79%) and cymol (14.73%) were high in cumin essential oil (Table 3). Antibacterial, antifungal and antioxidant activities of *C. cyminum* essential oil have been demonstrated (Bokaeian et al., 2014; Ghasemi et al., 2018). Studies have shown parallelism with our results and it has been reported that cumin essential oil contains high levels of cuminaldehyde compounds (Morcia et al., 2012). Khosravi et al. (2015) determined that *F. verticillioides* isolates were sensitive to *C. cyminum* essential oil and there was a significant decrease in the growth of fungal isolates. In the study, *C. cyminum* came to the forefront as the essential oil that inhibits fungus growth at the highest rate among 5 different essential oils. The antioxidant properties of sandalwood (*A. andrachne* L.), which belongs to the Arbutus genus of the Ericaceae family, are known. The main components found in sandalwood oil were cedrene (18.63%), eucalyptol (1.8-Cineole) (15.10%) and ethyl phthalate (7.23%) (Table 3). In the literature review, no study was found that investigated the antifungal effect of *A. andrachne*. This is the first study to investigate the antifungal effect of *A. andrachne*. According to the results we obtained, cedarwood essential oil inhibited the mycelial growth of *F. verticillioides* and *F. pseudograminearum* pathogens at varying rates depending on the dose. Ginger essential oil, which is an important spice plant from the Zingiberaceae family, contains high levels of sesquithujene (17.72%), eucalyptol (16.27%) and limonene (12.72%) (Table 3). In a study where ginger essential oil was applied against *F. verticillioides*, it was determined that ginger essential oil had a high antifungal effect against *F. verticillioides* (Castro et al., 2020).

Table 4. Antifungal properties of some plant essential oils on growth of *F. verticillioides and F. pseudograminearum* mycelium

Essential oils	Doses ($\mu L\ mL^{-1}$)	*F. verticillioides*		*F. pseudograminearum*	
		Mycelial growth (mm)[1]	MGI (%)	Mycelial growth (mm)[1]	MGI (%)
R. officinalis	0	43.2 a*	0.0	42.4 a*	0.0
	0.25	34.8 b	19.3	35.8 b	15.7
	0.5	30.1 c	30.3	30.9 c	27.1
	1	25.3 d	41.4	26.4 d	37.7
	2	21.2 e	50.9	22.6 e	47.4
	4	15.2 f	64.8	16.7 f	60.6
	Positive control	7.9 g	81.7	8.3 g	80.4
	$CV_{(0.01)}$	1.5		2.1	
N. sativa	0	43.0 a	0.0	38.8 a	0.0
	0.25	34.5 b	19.8	33.0 b	15.0
	0.5	29.3 c	31.9	27.7 c	28.7
	1	24.4 d	43.3	23.2 d	40.3
	2	19.9 e	53.7	19.1 e	50.8
	4	11.8 f	72.5	14.5 f	62.7
	Positive control	8.4 g	80.4	10.5 g	73.0
	$CV_{(0.01)}$	1.9		1.8	
C. cyminum	0	44.7 a	0.0	43.1 a	0.0
	0.25	38.2 b	14.5	37.1 a	13.9
	0.5	33.1 c	25.9	32.4 c	24.8
	1	26.7 d	40.2	27.5 d	36.2
	2	19.2 e	57.1	20.8 e	51.6
	4	11.3 f	74.6	13.7 f	68.3
	Positive control	8.0 g	82.1	9.1 g	78.9
	$CV_{(0.01)}$	6.1		3.4	
A. andrachne	0	43.9 a	0.0	41.9 a	0.0
	0.25	36.8 b	16.3	35.8 b	14.5
	0.5	33.2 c	24.4	32.4 c	22.7
	1	29.2 d	33.5	28.7 d	31.5
	2	24.5 e	44.2	24.3 e	42.0
	4	21.3 f	51.5	21.4 f	48.9
	Positive control	16.8 g	61.9	17.6 g	58.0
	$CV_{(0.01)}$	2.2		2.3	
Z. officinale	0	43.2 a	0.0	43.2 a	0.0
	0.25	34.3 b	20.5	35.3 b	18.3
	0.5	31.1 c	28.0	31.7 c	26.6
	1	25.3 d	41.4	26.9 d	37.7
	2	21.3 e	50.6	22.4 e	48.1
	4	14.6 f	66.2	16.6 f	61.6
	Positive control	8.8 g	79.6	10.9 g	74.7
	$CV_{(0.01)}$	2.3		3.0	

[1] Ten days after inoculation, the mean radial mycelial growth of *F. verticillioides* and *F. pseudograminearum* were calculated. Based on three replicate plates, each observation. Prior to statistical analysis, arcsine transformation was done.

*Mean values followed by different letters within the column are significantly different according to Duncan Test (P≤0.01). Positive Control: 80 g l^{-1} Triticonazole+40 g l^{-1} Pyraclostrobin-BASF company. MGI: Mycelial growth inhibition rate.

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4. Conclusions

Among the essential oils of rosemary, black cumin, cumin, sandalwood and ginger, which were found to have inhibitory effects in the study carried out under *in vitro* conditions, especially cumin essential oil was found to be the most effective essential oil against *F. verticillioides* and *F. pseudograminearum*. The highest inhibitory effect against both pathogens was obtained from positive control and high dose (4 µl mL^{-1}) treatment of cumin essential oil. The effect of synthetic fungicides used in the control against stem and ear rot disease decreases over time, resistance emerges, and human and environmental health are damaged. For this reason, detailed studies should be carried out to determine the *in vivo* activities of cumin and other plant essential oils, which can be used as an alternative to chemical pesticides, which do not disturb the natural balance, are considered safe for human and environmental health, and can be used as an alternative to chemical pesticides.

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Author contributions

Yagmur Ceylan: Data Curation/Visualization;
Zehra Saglan: Investigation/Data Curation;
Gurbet Celik Turgut: Resource/Instrument Supply/Review and Editing;
Oktay Erdogan: Methodology/Material/Original Draft Writing/Statistical Analysis/Supervision.

Conflict of interest

As the authors of this study, we declare that we do not have any conflict of interest statement.

Ethics Committee Approval

As the authors of this study, we declare that we do not have any ethics committee approval.

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