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INHIBITORY EFFECTS OF ESSENTIAL OILS OF SOME WILD MEDICINAL PLANT SPECIES ON ACTIVITY OF SOME PATHOGENIC FUNGI

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ABSTRACT

This study was carried out during 2013 – 2014 years to determine essential oils properties of some wild medicinal plant species [(Quysum) Achillea fragrantissima, (Zoeitran) Thymus decussates, (Samwah) Cleome droserifolia and (Myrrh) Tanacetum sinaicum)] and their antifungal inhibitory effects on four pathogenic fungi namely Alternaria alternata, Aspergillus niger, Aspergillus flavus and Fusarium oxysporum. The main components of Thymus decussates and Achillea fragrantissima oils were Thymol, Terpine-4-ol and Linolool alcohol. But, Cleome droserifolia oil was contain (E)-3,7,11-trimethyl , Cartotol , β -Eudesmol and Z-Myroxide. As for Tanacetum sinaicum oil, its main components were Thymol, Trans – Thujone and Trans-chrysanthenyl acetate. The antifungal activities of these oils at concentrations of 0.1, 1.3, 1.6 and 2.0 mg/ ml were investigated using disc-diffusion method. Also, the minimum inhibitory concentrations (MIC) of the 4 essential oils were determined by subjecting the tested fungi to different tested essential oils at concentrations ranging between 0.2 to 2.3 mg/ ml (21 treatments for each plant).

The low concentration (0.1 mg/ ml) of all tested essential oils did not show any inhibitory effects on the four examined fungi. At the same time, Thymus decussates essential oil at 2.0 mg/ ml followed by Achillea fragrantissima oil at 2.0 mg/ ml were the superior in their suppression on fungal activity of all tested fungi.

Cleome droserifolia essential oil had the highest inhibition on growth of Aternaria alternate, Aspergillus niger and Aspergillus flavns fungi when it used at concentration 2.0 mg/ ml. While, it inhibited Fusarium oxysporum growth when it used at 1.3 mg/ ml level. For Tanacetum sinaicum essential oil, its inhibitory effect on *all examined fungi was exhibited only when it used at moderate concentration of 1.3 mg/ ml.*

Respecting fungi sensitivity to essential oils, the minimum inhibitory concentrations test reveal that Aternaria alternata fungus followed by Aspergillus niger were the sensitive fungi to various tested essential oils. But, Aspergillus flavns was the resistant one and Fusarium oxysporum had moderate degree in its resistance against the tested essential oils.

***In conclusion:** results of such research confirm possibility of using the extracted natural essential oils of some medicinal plants as substitutes to chemical fungicides for overcoming the pathogenic fungi hazard effects.*

Key words: Antifungal, Minimum inhibitory concentration, Disk diffusion.

INTRODUCTION

Tremendous numbers of the Egyptian wild plants are documented in numerous reference books, researches and scientific reports. Too many wild plants showed medical effects, whether as sedatives, anti-bacteria and/ or anti-fungi. It was and still the desert population uses the wild plants in the treatment of most illnesses and in their food.

Many plants have limitless ability to synthesize aromatic substances of different functional groups, most of which are phenols or their oxygen substituted derivatives. Most are secondary metabolites, of which at least 13,000 have been isolated that is less than 10% of the total. In many cases, these substances serve as plant defense mechanisms against pathogenic microorganisms, insects, and herbivores. Some plants used for their odors (terpenoids), pigment, (quinones and tannins) and/ or flavor (terpenoid capsaicin from chili peppers) were found to be endowed with medicinal properties. Some of the herbs and spices used by humans as season food yield useful medicinal compounds (Cowan, 1999).

Quysum *(Achillea conferta* Forssk. Fam. *Asteraceae*) is used practically for many medical purposes, but it is considered as less strong because its smell is less intense. According to direct field experience, Bedouins use *Achillea santolina* for medicinal purposes; also this species has fewer effects than *Achillea fragrantissima* (Ozenda, 1991).

Zoeitran (*Thymus decussates* Benth. Fam. *Labiatae*) is one of the world's more well-known and useful plants. Its fresh and dry leaves are used for flavoring food, in addition the essential oil adding many medicinal values. It has been used since ancient times for its culinary, medicinal properties, and even presumed magical qualities. Ancient Egyptians used thyme in embalming. Also, the ancient Greeks used it in their baths and burnt it as incense in their temples, believing that thyme was a source of courage. Courage is *"thumus"* in the Greek language, and may be the inspiration for the generic name. (Hanrahan and Odle, 2005).

Samwah (*Cleome droserifolia* Forssk. Fam. *Capparaceae*) is a rare desert species in Egypt. This plant goes up to the limits of a meter and has green leaves and twigs characterized by greenery dark near the lemon leaves color. This plant is one of the most important plants in the treatment of diabetes, sores, wounds and loss of appetite (Hanrahan and Odle, 2005).

Myrrh *(Tanacetum sinaicum* Feinbr. Fam. *Asteraceae*) is small thorny tree species. It produces aromatic resin containing an essential oil termed *oleoresin*. Myrrh resin is a natural gum. It has been used throughout history as a perfume, incense and in medicine. It can also be ingested by mixing it with wine. Myrrh is used more frequently in Ayurveda and Unani medicine, which ascribe tonic and rejuvenative properties to the resin. It is utilized in many specially processed *rasayana* formulas in Ayurveda. However, non-*rasayana* myrrh is contraindicated when kidney dysfunction or stomach pain is apparent, or for women who are pregnant or have excessive uterine bleeding (Morrow 2011).

Molds and fungi are very common in our environment, and there occurrence frequently caused many plant diseases and food contaminations. Many chemicals were used to control growth of mold and fungi. But, some of these substances may have offensive odors and others cannot be added to plants and food for their hazardous effects.

Generally, phytopathogenic fungi are controlled by synthetic fungicides; however, use of these fungicides is increasingly restricted due to their harmful effects on human health and the environment (Harris *et al*., 2001). Increasing demand of production and regulation on the use of agrochemicals and the emergence of pathogens resistant to the products employed justifies the search for novel active molecules and new control strategies. Since antiquity, plants from the genus *Achillea and Thymus* grown in Sant-Kathrin in Egypt and has provided a variety of compounds of known therapeutic properties, like analgesics, anti-inflammatories, medicines for asthma, and others. In recent years, antimicrobial properties of plant extracts have been reported with increasing frequency from different parts of the world (Cowan, 1999). Plants from the genus, *Cleome and Tanacetum* are commonly grow wildly in Sinai and used in veterinary medicine in latten America and Africa to treat animal problems popularly diagnosed as "mycoses" (Demo and Oliva, 2008). Several works have demonstrated in laboratory trials that different plant tissues, such as roots, leaves, seeds and flowers possess inhibitory properties against bacteria, fungi and insects (Davicino *et al*., 2007).

Fungi are ubiquitous in the environment, and infection due to fungal pathogens has become more common. The genus *Alternaria, Aspergillus* and *Fusarium* are widely distributed in nature and its species are among the most common fungi on the philosophers (Lopes and Martins, 2008). It includes both plant-pathogenic and plant-saprophytic species that may damage crops in the field or cause post-harvest decay (Griffin and Chu, 1983), causing considerable economics losses for farmers and food industries. In addition, the genus produces mycotoxins and phytotoxins, and studies in the last decade have emphasized its toxicogenic properties rather than simply those that cause spoilage.

In world crop production, pre harvest losses due to fungal diseases may amount to 12 % in developing countries. Using chemicals for fungal diseases control pollute the atmosphere and adversely affect the properties of medicinal plants. To avoid the hazardous effects of chemicals, natural products of some plants have been used to control plant diseases (Bowers, and Locke, 2000; Momin and Nair, 2001).

Fungal species of genera *Aspergillus, Fusarium, Alternaria* and *Drechslera* species have been considered to be major plant pathogens worldwide (Mirza and Queshi, 1982). *Aspergillus* and *Fusarium* species produce mycotoxins which contaminate foods and cause many adverse effects on human and animals' health fed on such contaminated agricultural products, besides they cause seedling blight; seed, kernel, and stalk rot and plant stunting (Fandohan *et al*., 2003).

According to the available literature there is little information about the inhibitory effect of volatile oils on pathogenic fungi growth.

So, such work aimed to evaluate the antifungal properties of essential oils belonging to four medicinal plants (*Achillea fragrantissima, Thymus decussates*, *Cleome droserifolia* and *Tanacetum sinaicum*) grown wildly at Santa-Katherine Protectorate, South Sinai governorate, Egypt against four pathogenic fungi namely *Alternaria alternata, Aspergillus niger, Aspergillus flavus* and *Fusarium oxysporum*.

MATERIALS AND METHODS

This study was carried out during 2013 – 2014 years at Hort. Res. Inst. Lab., Food Tech. Res. Inst. Lab., and Central Agric. Pesticide Lab. Agric. Res. Center, Giza, Egypt aiming to assess essential oils properties of some wild medicinal plant species and their antifungal inhibitory effects on four pathogenic fungi.

Gathering plants and extraction of their essential oils:

Four medicinal plants; *i.e.,* Quysum (*Achillea fragrantissima*), Zoeitran (*Thymus decussates*), Samwah (*Cleome droserifolia*) and Myrrh (*Tanacetum sinaicum*) grown wildly at Sant-Katherine Protectorate were collected with the participation of the Ministry of Environmental Affairs, South Sinai - South Sinai governorate, Egypt. Botanical classification of collected plants has been confirmed according to the Environmental Affairs Department, plant flora. The herb were subjected to air draying in good airy and shady place for 10 days, then essential oils samples of the abovementioned collected plants were extracted. Essential oil extraction was done using steam distillation in Clevenger apparatus for three hours. Extracted oil samples were dried using anhydrous sodium sulphate, then stored at 4°C in dark (British Pharmacopoeia, 1963). Also, essential oils percentages in herb were calculated on dry weight bases and were expressed as mean of three samples (Table 1).

Table 1: Essential oils percentages and their physical properties of *Achillea fragrantissima*, *Thymus decussates*, *Cleome droserifolia* and *Tanacetum sinaicum* plants.

Volatile oil source	Essential oil In dried herb (%)	Specific gravity (15^{o}C)	Refractive index (20^{o}C)	Optical rotation
Achillea fragrantissima	1.20	0.925	1.4886	+1^o
Thymus decussates	2.60	0.911	1.4925	+1^o
Cleome droserifolia	1.10	0.901	1.4770	+ 1^o
Tanacetum sinaicum	0.60	0.988	1.4878	+ 0.8^o

Physical properties of essential oils under study:

Specific gravity, refractive index and optical rotation were determined in essential oils samples which extracted from the 4 abovementioned medicinal plants using the methods described by A.O.A.C. (1995). The determined properties of *Achillea fragrantissima*, *Thymus decussates, Cleome droserifolia* and *Tanacetum sinaicum* essential oils are recorded in Table 1. However, most values were found to be within the range which previously noticed by Guenther (1982).

Main components of essential oils under study:

The main components of the four essential oils of *Achillea fragrantissima, Thymus decussates, Cleome droserifolia* and *Tanacetum* *sinaicum* were determined. GC-MS analysis was conducted on a Hewlett-Packard 5985 coupled with a HP MS instrument system. The ionization voltage was 70 EV (Electric Volt) and the ion source temperature was 200 ^{o}C. Components were identified using the (NBS) MS library or other published mass spectra and by comparing their retention index with the published data (Robert Adams 1995). GLC analyses of the essential oils showed that *Thymus decussates* and *Achillea fragrantissima* oils were characterized by the presence of oxygenate compounds such as Thymol, Terpine-4-ol and Linolool alcohol. But, *Cleome droserifolia* oil was characterized by the presence of (E)-3,7,11-trimethyl, Cartotol, β -Eudesmol and Z-Myroxide. As for *Tanacetum sinaicum* oil, its main components were Thymol, Trans – Thujone and Trans-chrysanthenyl acetate (Table 2).

Microbial cultures sources:

Microbial cultures of *Aspergillus niger* and *Aspergillus flavus*, were kindly obtained from Faculty of Agriculture, Ain Shams University, Egypt. While, *Alternaria alternate* and *Fusarium oxysporum* cultures were kindly supplied from Central Agriculture Pesticide Lab., Agriculture Researh Center, Giza, Egypt.

Recorded data:

Essential oils inhibitory effects on activity of the examined fungi were evaluated via two means as follows:

a) ***Determination of inhibition zones (mm) of different fungi under the effect of tested essential oils:***

Disc diffusion method described by **Yin and Tsao (1999**) was used to determine the efficiency of the tested essential oils on inhibition the examined fungi activity. In brief, sterile nutrient agar at 43-45 ^{o}C was poured into Petri plates (9 cm diameter), then it was solidified at 4 ^{o}C for *1hr*, and then 0.2 ml of each culture organism (*Alternaria alternata, Aspergillus niger, Aspergillus flavus and Fusarium oxysporum*) inoculum was inoculate per Petri plate. Inoculum was eventually spread on agar using a glass rod spreader. The Petri plates were left at 4 ^{o}C for 1 *hr* to allow agar surface to solidify. Sterile filter papers 16 mm diameter were placed on the culture media and were impregnated with different essential oils at concentrations of 0.1, 1.3, 1.6 and 2.0 mg/ ml for each. Impregnation was done by placing essential oils on the culture media and on the inoculated plates. Plates were turned upside down and incubated at 30 ^{o}C for 24 *hr*. At the end period, the formed inhibition zones in each culture were measured in millimeters.

Table 2: Chemical composition of essential oils of *Achillea fragrantissima*, *Thymus decussates*, *Cleome droserifolia* and *Tanacetum sinaicum* plants.

The component (%)	Essential oil of:			
	Achillea fragrantissima	*Thymus decussates*	*Cleome droserifolia*	*Tanacetum sinaicum*
α- Pinene	1.45	1.40	-	1.49
β-Pinene	4.55	0.30	-	1.39
Terpine-4-ol	15.7	--	-	1.86
Verbenone	4.42	--	-	--
α- Terpinene	2.40	0.90	-	0.67
Linolool Thymol Terpine -4-ol	11.0	4.70	-	0.59
Carvone	9.42	--	-	-
γ-terpinene	5.60	--	-	-
Myrcenol	1.40	0.70	-	-
P- cymene	0.25	0.60	-	0.45
Eugenol	-	--	-	4.55
D-terpinene	-	--	-	-
Thymol	--	26.3	-	17.9
Thymol methyl ether	--	10.8	-	--
ß-Caryophyllene	--	7.50	-	--
p-Cymene	--	19.9	-	0.46
(E)-3,7,11-trimethyl Cartotol β -Eudesmol Z-Myroxide	--	--	11.9	--
Cartotol	--	--	10.1	--
σ–Cadinene	--	--	8.88	--
β -Eudesmol	--	--	6.91	--
benzyl isothiocyanate	--	--	4.90	--
Z-Myroxide	--	--	9.01	--
3,5-dimethyl-1,2,4-trithiolane	--	--	7.06	--
Trans – Thujone	--	--	--	17.5
Trans-chrysanthenyl acetate	--	--	--	13.2
Umbellulone	--	--	--	9.74

b) Determination minimum inhibitory concentrations (MIC) of essential oils under study on the tested fungi:

For assess the MIC of each examined essential oil on different tested pathogenic fungi, sterile 5 ml of each essential oil at different concentrations of 0. 2, 0.3, 0.4..… to 2.3 mg/ ml were poured into sterile empty tubes and then l ml of each organism inoculum was added to each tube. After that, l ml of (essential oil + organism) culture was added onto 5ml of sterile nutrient agar in tubes. Then, the tubes were incubated at 30 ^{o}C for 15 days. Eventually, observations were noticed for visible growth of fungi. The highest dilution (lowest concentration) which began to inhibit the fungal growth was detected and it was represented the MIC (Rasooli and Abyaneh, 2004 and Ramzy *et al*., 2009).

Statistical Analysis

Determinations of essential oils percentages and the antifungal effect of the essential oils under study in this research were repeated three times, and the results were expressed as means of the three values of each recorded determinations.

RESULTS AND DISCUSSION

Antifungal effect of the essential oils under study:

a. ***Inhibition zones (mm) of different fungi under the effect of tested essential oils:***

Results of inhibitory effects determined as diameter of inhibition zones (mm) which resulted from adding different essential oils under study at different tested concentrations on the examined fungi cultures are shown in Table 3.

Recorded data indicate that all the tested essential oils had antifungal effects against the four tested fungi. Different responses were noticed in the inhibition effects related to different oils and different fungi. The low concentration (0.1 mg/ ml) of all tested essential oils did not show any inhibitory effects on the four examined fungi.

In general, *Thymus decussates* essential oil at 2.0 mg/ ml followed by *Achillea fragrantissima* oil at 2.0 mg/ ml recorded very powerful antifungal activity towards all tested fungi as compare to the other tested essential oils.

As for *Cleome droserifolia* essential oil inhibition effects, data of the same Table 3 show that it had the highest inhibition on growth of *Aternaria alternate*, *Aspergillus niger* and *Aspergillus flavns* fungi when it used at concentration 2.0 mg/ ml. While, *Fusarium oxysporum* fungus growth was inhibited under *Cleome* essential oil effect at 1.3 mg/ ml concentration.

Tanacetum sinaicum essential oil inhibitory effect on all examined fungi was exhibited only when it tested at moderate concentration of 1.3 mg/ ml. Decrease or increase its concentration (0.1 or 1.6 mg/ ml, respectively) had no effect in this respect. Also, its high tested concentration

Table 3: Diameter of inhibition zones (mm) resulted by essential oils of *Achillea fragrantissima*, *Thymus decussates*, *Cleome droserifolia* and *Tanacetum sinaicum* plants on *Alternaria alternate, Aspergillus niger, Aspergillus flavns* and *Fusarium oxysporum* fungi

-The fungus / Essential oil	***Alternaria alternate***	***Aspergillus niger***	***Aspergillus flavns***	***Fusarium oxysporum***
Essential oils concentration at 0.1 mg/ ml				
Achillea fragrantissima	☺NIZ	NIZ	NIZ	NIZ
Thymus decussates	NIZ	NIZ	NIZ	NIZ
Cleome droserifolia	NIZ	NIZ	NIZ	NIZ
Tanacetum sinaicum	NIZ	NIZ	NIZ	NIZ
Essential oils concentration at 1.3 mg/ ml				
Achillea fragrantissima	NIZ	NIZ	NIZ	NIZ
Thymus decussates	NIZ	NIZ	21	NIZ
Cleome droserifolia	22	NIZ	NIZ	NIZ
Tanacetum sinaicum	NIZ	NIZ	NIZ	NIZ
Essential oils concentration at 1.6 mg/ ml				
Achillea fragrantissima	27	28	21	22
Thymus decussates	29	28	24	26
Cleome droserifolia	25	24	NIZ	NIZ
Tanacetum sinaicum	NIZ	NIZ	NIZ	NIZ
Essential oils concentration at 2.0 mg/ ml				
Achillea fragrantissima	31	32	25	26
Thymus decussates	33	32	27	29
Cleome droserifolia	29	28	19	NIZ
Tanacetum sinaicum	NIZ	24	NIZ	NIZ
Essential oils concentration at 2.4 mg/ ml				
Achillea fragrantissima	31	32	25	26
Thymus decussates	33	32	27	29
Cleome droserifolia	29	28	19	24
Tanacetum sinaicum	29	27	21	22

☺NIZ =No. of inhibition zone

of 2.0 Mg/ ml recorded more inhibition effect on *Aspergillus niger* fungus as compare to the moderate concentration of 1.3 mg/ ml (Table 3).

Also, data reveal that *Aternaria alternata* fungus followed by *Aspergillus niger* were the sensitive fungi to various essential oils comparing to the other tested fungi. But, *Aspergillus flavns* was the resistant one against the tested essential oils.

The antifungal activity of essential oils against micro-organisms might be due to the presence of phenolic terpenes that cause damage to the biological membrane due to lipophilic properties interfering with membrane-integrated enzyme (El-Baroty (1988). Volatile oil plants have been generally recognized as safe (Newberne *et al*., 2002). Systematic screening for biological interactions between microorganisms and plant products has been valuable source of new and effective antimicrobial substances, which could have different action ways on microbial cell when compared to other conventional antimicrobials plant synthesized by a secondary metabolism. These compounds with complex molecular structures and some of them have been related with antimicrobial properties found in plant and their derivatives. Among these secondary metabolites are found trepenes and phcnolic compounds (Simoes *et al*., 1999).

b. The minimum inhibitory concentrations (MIC) of essential oils under study:

Results in Table 4 indicate that *Thymus decussates* oil had the least value of MIC for all tested fungi compared with other essential oils followed by *Achillea fragrantissima*, *Cleome droserifolia* and *Tanacetum sinaicum*, respectively which indicated that Zoeitran and Quysum essential oils had a powerful antimicrobial activity against all the tested fungi. Indicating that there is a relationship between the chemical composition of the oil and its powerful inhibitory against microorganisms.

This inhibitory effect might be due to the presence of group which is more active and easily forms bonds with the active sites of some organism enzymes (El-Baroty 1988). Zoeitran oil had the highest antifungal activity due to oxygenate phenolic terpenes which is a derivative of thymol and p-Cymene. It is very inhibitory against *Aspergillus niger*. Naganawa *et al*. (1996) and Yoshida *et al*., (1987) reported that oxygenate phenolic terpenes have stronger antifungal activity. They reported that phenolic terpenes damage the cell walls of fungi.

Quysum essential oil show strong inhibitory effects against *Aspergillus niger* (Benkeblia, 2004). The powerful antifungal activity of Quysum essential oil might be due to Terpine-4-ol and Carvone component, an antifungal compound isolated from quysum, since this compound shows antifungal activities against several fungal species (Phary *et al*., 1999).

Benkeblia (2004) observed inhibitory effects of some oxygenate components in some essential oils on *Aspergillus niger* and *Fusarium oxysporum*. As for Myrrh oil, its antifungal activity may be due to Thymol and Eugenol components.

Many authors have emphasized that the antimicrobial effect of essential oil constituents has been dependent on their hydrophobicity and partition in the microbial plasmatic membrane. Effect of specific ions due to their addition in/ on plasmatic membrane had great effect on the protons motive force and overall activity of microbial cells, including tug or pressure control, solutes transport and metabolism regulation (Lanciotti

Table 4: Minimum inhibitory concentration (MIC mg/ ml)) of the test essential oils on growth of *Alternaria alternate, Aspergillus niger, Aspergillus flavns* and *Fusarium oxysporum* fungi

Volatile oil source	The fungus:			
	Alternaria alternate	***Aspergillus Niger***	***Aspergillus flavns***	***Fusarium oxysporum***
Achillea fragrantissima	0.8	0.7	1.3	1.1
Thymus decussates,	0.4	0.6	0.8	0.7
Cleome droserifolia	1.3	1.2	1.9	2.2
Tanacetum sinaicum	2.4	1.7	2.2	2.3

et al., 2004). Antifungal activity of Zoeitran essential oil may be attributed to the presence of thymol and other terpene compounds which have synergistic effects against fungi. Also, results of this research were in agreement with those reported by El-Baroty (1988), Mousa (1998) who observed that there is a relationship between the antifungal activity of the volatile oils and its chemical composition.

Conclusively, results of such research confirm possibility of using the extracted natural essential oils of some medicinal plants as substitutes to chemical fungicides for overcoming the pathogenic fungi hazard effects. However, more researches are needed to determine the specialty and specific concentrations of essential oils on different fungi.

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التأثيرات المثبطة للزيوت العطرية لبعض أنواع النباتات الطبية البرية على نشاط بعض الفطريات الممرضة

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أجريت هذه الدراسة خلال عامي ٢٠١٣- ٢٠١٤ لتقدير خصائص الزيوت العطرية لبعض أنواع النباتات الطبية البرية [(القيسوم) *Achillea fragrantissima* ، (الزعتران) *Thymus decussates* ، (الصموة) *Cleome droserifolia* ، (المر) *Tanacetum sinaicum*] وتأثيرات زيوتهم العطرية المثبطة على نشاط اربع فطريات ممرضة أي : الألترناريا *Alternaria alternate* ، أسبرجلس نيجر *Aspergillus niger* ، أسبرجلس فلافس *Aspergillus flavus* ، فيوزاريوم *Fusarium oxysporum* .

كانت المكونات الأساسية لزيتي نباتي الزعتران والقيسوم هي Thymol ، Terpine-4-ol ، Linolool alcohol ، بينما احتوى زيت نبات الصموة على (E)-3,7,11-trimethyl ، Cartotol ، β –Eudesmol ، Z-Myroxide ، أما زيت المر فكانت مكوناته الأساسية هي الـ Thymol ، Thujone and Trans-chrysanthenyl acetate.

فُحصت الأنشطة المضادة للفطريات لهذه الزيوت العطرية عند تركيزات ٠.١ ، ١.٣ ، ١.٦ ، ٢.٠ مجم/ ملليلتر باستخدام طريقة الانتشار في الأقراص disc-diffusion method ، أيضاً قُدر أقل تركيزات مثبطة للأربع زيوت العطرية عن طريق تعريض الفطريات المختبرة لمختلف للزيوت العطرية بتركيزات تتراوح بين ٠.٢ إلى ٢.٣ مجم/ ملليلتر (٢١ تخفيف).

لم يظهر التركيز المنخفض (٠.١ مجم/ ملليلتر) لكل الزيوت العطرية المختبرة أي تأثيرات مثبطة على الأربع فطريات المختبرة ، في نفس الوقت كان زيت الزعتران بتركيز ٠.٢ مجم/ ملليلتر تلاه زيت القيسوم بتركيز ٠.٢ مجم/ ملليلتر الأكثر تثبطاً للنشاط الفطري لكل الفطريات المختبرة.

امتلك زيت الصموة أعلى تثبيط على نمو كل من فطريات الألترناريا *Aternaria alternate* ، أسبرجلس نيجر *Aspergillus niger* ، أسبرجلس

فلافس *Aspergillus flavns* عند استخدامه بتركيز ۰.۲ مجم/ ملليلتر ، بينما ثبط نمو فطر الفيوزاريوم *Fusarium oxysporum* عند استخدامه بمستوى ۱.۳ مجم/ ملليلتر ، بخصوص زيت المر فقد ظهر تأثيره المثبط على كل الفطريات المختبرة فقط عند استخدامه بالتركيز المتوسط ۱.۳ مجم/ ملليلتر.

بخصوص حساسية الفطريات الأربع للزيوت العطرية المختبرة ... عكس اختبار أقل تركيز مثبط أن فطر الألترناريا *Aternaria alternate* تلاه فطر أسبرجلس نيجر *Aspergillus niger* كانا الفطريات الأكثر حساسية لمختلف الزيوت المختبرة ، بينما كان فطر أسبرجلس فلافس *Aspergillus flavns* الأكثر مقاومة ، وامتلك فطر الفيوزاريوم *Fusarium oxysporum* درجة متوسطة من المقاومة للزيوت العطرية المختبرة.

التوصيةً: أيدت نتائج هذا البحث إمكانية استخدام الزيوت العطرية الطبيعية المستخلصة من بعض النباتات الطبية كبدائل للمبيدات الفطرية الكيماوية من أجل التغلب على الآثار الضارة للفطريات الممرضة.

المحكمون:

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