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EFFECT OF SOME PLANT EXTRACTS AND ESSENTIAL OILS ON FABA BEAN WILT CAUSED BY FUSARIUM OXYSPORUM F.SP. FABAE

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ABSTRACT: *Fusarium oxysporium* f. sp. *fabae* (FOF) the causal organism of faba been wilt were the most frequently isolated fungi from Menouf, Sadat City, El-Bagour, Quesna, Tala and Shebin El-kom, localities of Menoufia governorate. pathogenicity test experiment proved that the most aggressive isolate of *Fusarium oxysporium* f.sp. *fabae* (FOF1) was achieved from Menouf district. Three faba bean lines; *i.e* Australia, Balady and English were evaluated against the most virulent isolate under greenhouse conditions. In this study, six plant extracts were used (Mint, Clove, Cactus, Cinnamon and, Camphor) in concentrations 5, 10 and 15 %), and six essential oils (Clove, Cinnamon, Garlic, Thyme and, Sesame) in concentrations of 2.5, 5 and 10 %) These plant extracts and essential oils were tested on the inhibition, growth, and sporulation of (FOF). Both under laboratory and green house conditions. pathogenicity test experiments proved that the most aggressive isolate of (FOF) was that achieved from Menouf district (FOF1). Under laboratory conditions plant water extracts and plant essential oils individually reduced the average linear growth of (FOF) and spore production. Clove and cinnamon plant extracts showed the best efficiency. While Cinnamon essential oil gave best effect even at lowest concentration. However, under green house and artificially soil infestation conditions, the application of either plant extracts and plant essential oils decreased the disease incidence, improved faba bean plant growth and reduced the spore population in the soil; significantly.

Key words: *Fusarium oxysporium*, faba bean wilt, plant extract, and essential oils.

INTRODUCTION

Legume crops, a sustainable source of high-protein food, they are grown widely world. Among legumes, faba bean (*Vicia faba* L.), is one of the oldest crops cultivated worldwide (Mínguez and Rubiales, 2021). Many countries such as Ethiopia, Egypt, China, Afghanistan, India, Northern Europe, and Northern Africa, are major producers of faba bean (Rahate *et al*., 2020). Approximately 2.5 million ha of faba bean are planted in the world (Wang *et al*., 2014) Faba bean (*Vicia faba* L.) is one of the most important legume crops mainly grown as a valuable protein-rich food, both for human and animals' consumption and has 7.7% carbohydrates that human needs daily Vasic *et al*., (2019). This crop is so important to the soil, where it gives the soil about 150-200 nitrogen unit.

Faba bean is vulnerable to many phytopathogenic fungi that can have a significant impact on reducing its yield and quality, Fusarium wilt one of the most common and destructive soil-borne fungal disease of faba bean because of continuous monocropping. It has been responsible for severely reduction yield which causes serious economic losses worldwide and limits faba bean production (Stoddard *et al*., 2010).

Antimicrobial chemicals such as fungicides are often used in control of plant diseases. Two major concerns about chemical residue in the environment and the development of resistance by the pathogen (Spotts and Cervantes 1986; Osuinde *et al*., 2001). Under this concept, all possible trails of plant pests and disease control methods should be integrated to minimize the excessive use of synthetic pesticides. In the few decades, the scientists go to search about nonchemical approaches to diseases control (Wilson *et al*., 1987).

To avoid the use of chemical fungicides many of the eco-friendly compounds could be used such as plant extracts. Several plants extracts have been found to possess outstanding fungi toxicity against mycelial growth and spore

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germination of different phytopathogenic fungi in vitro (Tripathi *et al*., 2008). Also plant essentials oils, have been examined its effect on fungi to search for natural fungicides and numbers of these oil constituents have shown inhibitory effects (Chao *et al*., 2000). the use of the plant extracts and essential oils is recently advocated by several research, as a potential control method of plant diseases.

The objectives of these studies were searching about an alternative control method for reducing the use of fungicides by using some plant extracts and essential oils as an antifungal activity, on (FOF) the causal organism of faba bean wilt.

MATERIALS AND METHODS

1. Isolation, purification, and identification of the tested microorganisms:

1.1. Pathogenicity test experiments:

Pathogenicity test experiments were carried out under greenhouse conditions at Faculty of Agriculture farms, Menoufia University, Shebin El-Komi, Egypt from 2018 to 2021 growing season. Clay loam soil was autoclaved twice at 121°C for one hour. Pots (20 cm in diameter) were sterilized by dipping in 5% formalin for 5 minutes and left for a week until formalin was evaporated. The isolated fungi were individually grown on Barley medium (75g barley grains + 25 g sand + 100 ml water); using 500 ml conical flasks. The flasks were incubated for 14 days at 25°C which were shacked every day to allow more fungal growth. Sterilized soil was infested separately with each isolate at the rate of 3% of soil weight. The infested soil was irrigated every day for a week to allow the fungus distribution into. faba bean (*Vicia faba*) seeds Cultivars (Australian, Balady and English) were separately surface sterilized by dipping into 1% formalin solution for 30 seconds, rinsing by sterilized distilled water and left to dry before sowing in the infested soil. Control treatment had the sterilized soil with the same percentage of sterilized Barley medium. three replicates were used for each treatment and the pots were irrigated as needed. The plants were examined every week for disease incidence determination.

2. Laboratory experiments:

A complete randomized design with three replicates was followed in these experiments.

2.1. Effect of plant extracts on the fungal growth and Sporulation:

Different preparations of plant extracts were used. All powder plant materials involved in this study, were collected, and identified (English name, scientific name, Arabic name and used parts) in Table (1). Two hundred grams of each tested plant powder were soaked in1000 ml sterilized distilled water. The obtained extracts were separately autoclaved at 90°C for 90 minutes, then filtered through filter paper, completed to be one liter (Awad and El-shennawy, 2015). Three concentrations of each aqueous extracts i.e., 5, 10 and 15 % were used. Extracts were prepared and evaluated for bioactivity by agar dilution method (De Rodrigues *et al*., 2005; Akaeze and Modupe, 2021).

Table (1): Medicinal and ornamental plants used for extraction.

Arabic name	English name	Scientific name	Used part
النعناع	Mint	*Mentha arvensis*	Leaves
القرنفل	Clove	*Syzygium aromaticum*	Fruits
الصبار	Cactus	*Aloe vera*	Stem and leaves
القرفة	Cinnamon	*Cinnamomum verum*	Leaves
الكافور	Camphor	*Cinnamomum camphora*	Bark

C1 * V1 = C2 *V2

C1: More concentrated solution V1: Volume needed for a more concentrated solution

C2: Final concentrated solution

V2: Desired volume for the final solution However, control treatment had PDA medium only.

2.2. Effect of some essential plant oils on the fugal growth and sporulation:

Essential oils of clove, garlic, thyme, sesame, and cinnamon were obtained from El - Gomhouria company for oils, Cairo, Egypt. The oils were emulsified with 3 % (v: v) tween 20. The emulsified oils were separately mixed with PDA medium to obtain the concentrations of 2.5, 5 and 10 % while control treatment received tween 20 at the some used concentration (Fontes *et al.*, 2018).

Different volumes of either essential oils and/or plant extracts were mixed with the sterile PDA to obtain various concentrations. The supplemented PDA dishes were inoculated with agar disc (5 mm in diameter) of FOF (from 8 - day - old PDA cultures). They were incubated at 25 ± 2 ° C for 8 days. Then the blocking fungal development was calculated (Ylar and Kadoglu. 2016).

3. Data recorded:

3.1. Growth diameter:

The average diameter of the fungal growth (mm) was recorded when a Petri dish of the experiment showed full growth. Percentage of inhibition over control was calculated as the formula of Sundaramoorthy and Balabaskar 2013:

$$PI\% = \frac{C - T}{C} X\ 100$$

PI = percent inhibition over control
C: Mycelial radial growth in control
T: Mycelial radial growth in treatment

3.2. Spore population:

The tested isolate of FOF was grown on potato - dextrose agar (PDA) in darkness at 22-25 °C for one week. By the aid of camel hairbrush, the formed spores were gently removed using 10 ml sterilized distilled water. Spore suspension was then, filtered through a layer of Mira cloth and the suspension was diluted and counted using haemo-cytometer (Jahanshir and Dzhalilov, 2010) at the end of the experiment. Finally, the average number of spores / ml was calculated according to the formula: No. of spore / ml = AV. No. of spores x dilution x slide factor

4. Greenhouse experiments:

Greenhouse experiments were carried out at the farm of Faculty of Agriculture, Menoufia University, Shebin El - Komi, Egypt; during 2018 and 2021 growing seasons. Pots and soil sterilization and soil infestation were conducted as mentioned in Pathogenicity test experiments.

Table (2): Essential oils used.

Arabic name	English name	Scientific name
القرنفل	Clove	*Syzygiumaro maticum*
القرفة	Cinnamon	*Cinnamomum verum*
الثوم	Garlic	*Allium sativum*
الزعتر	Thyme	*Thmus vulgaris*
السمسم	Sesseme	*Sesamum indicum*

4.1. Varietal resistance:

From the obtained results of pathogenicity test, the most virulent isolate of *Fusarium oxysporum* f. sp. *fabae* ((menouf isolate (FOF1)), was selected for this experiment. The sand / cornmeal / wheat bran medium (1: 2: 2 v / v) was backed in polyethylene bags, watered, and autoclaved for 30 minutes at 121°C. Then it was inoculated with equal disks of 7 days old mycelial growth of the above-mentioned selected isolate. The bags were incubated for 15 days at 28 °C. Three faba bean genotypes, lines, and hybrids; i.e Australia, Balady and English were evaluated against the most virulent isolate under greenhouse conditions. Faba bean seeds were surface sterilized as described in pathogenicity test. Five seeds were sown in each polyethylene pot and three replicates for each treatment were used. Surface sterilized seeds were sown in non-infested soil as control (three replicates). Polyethylene pots were kept in greenhouse; irrigate periodically and received all other normal agricultural practices. The obtained results were tabulated and statistically analyzed.

4.2. Effect of plant water extracts and essential oils on the pathogen population in the soil and faba bean growth parameters:

4.2.1. Effect on the pathogen population:

Faba bean (*Vicia faba*) seeds cultivar Australian were sown in the pots previously infested with (FOF1) (3 %), 5 seeds / pot; of soil weight ten days after soil infestation. The pots were irrigated by different water plant extracts at the concentrations of (5, 10 and 15%) (Table 1) and essential oils at the concentrations of (2.5, 5 and 10%) (Table 2) at the rate 75 ml / pot. However, control treatment pots received the same amount of sterilized distilled water instead of the extracts every week. At the end of the experiment, 10 grams of the middle of potted soil was picked up and added to 99 ml sterilized distilled water. Of this 1:100 dilution; the spores of *Fusarium oxysporum* were counted using Haemo-cytometer (Starovic *et al*., 2016). (1/400 m); The above - mentioned treatment methods were carried out from each replicate of each treatment. The same methods were followed for the efficacy of tested oils on the pathogen population into the soil. Emulsified oils were tested at 2.5, 5 and 10 % concentrations (Abhishek Sharma *et al.,* 2018).

4.2.2. Effect on the disease incidence:

Both percentage of infection and severity of infection with wilt disease were estimated after 55 days of sowing. Wilt disease percentage of infection (PI) was determined according to this formula:

$$PI = \frac{\text{No. of diseased plants}}{\text{Total No. of plants}} \times 100$$

However, severity of infection (SI) was estimated as the length of browning vesicles (cm).

4.2.3. Effect on plant growth arameters:

The average of plant height plants was determined in relation to the disease incidence at the end of the experiments.

5. Statistical analysis:

All experiments were conducted in completely randomized design. Mean values were compared by the least significant difference (LSD) testing at p= 0.05. Duncan's multiple Range test at p =0.05 was used to compare means. All statistical analyses were performed using Costate, Statistical Software.

RESULTS

1. Frequency of the isolated pathogens and biocontrol agents from different localities of Menoufia governorate:

Results present in Fig (1) clear that both (FOF) and *F.solani* were more frequently observed at El-Bagour district followed by Menouf one. They were less observed at Sadat City and Tala districts. However, both fungi were isolated from the samples obtained from all examined districts. *Rhizoctonia solani* was highly present in Sadat City samples, followed by those of Tala ones. The highest frequency of

Alternaria solani was achieved from Menouf and shebin EL-Kom regions. On the other hand, *Trichoderma harzianum* and *T. viride* isolates were more prevalent in Quesna, Sadat City and Menouf samples.

2. Pathogenicity test of *Fusarium oxysporum* isolates obtained from different localities of Menoufia governorate:

Results showed in Table (4) indicate that the most aggressive *Fusarium oxysporum* isolate was that obtained from Menouf district (F1) where it resulted the average of 4.67 collapsed Plant and only 0.33 survival ones. However, the least pathogenic *F. oxysporum* isolates were those achieved from Tala and shebin EL-Kom regions. Generally, all *F. oxysporum* isolates were pathogenic to faba bean Balady cultivar.

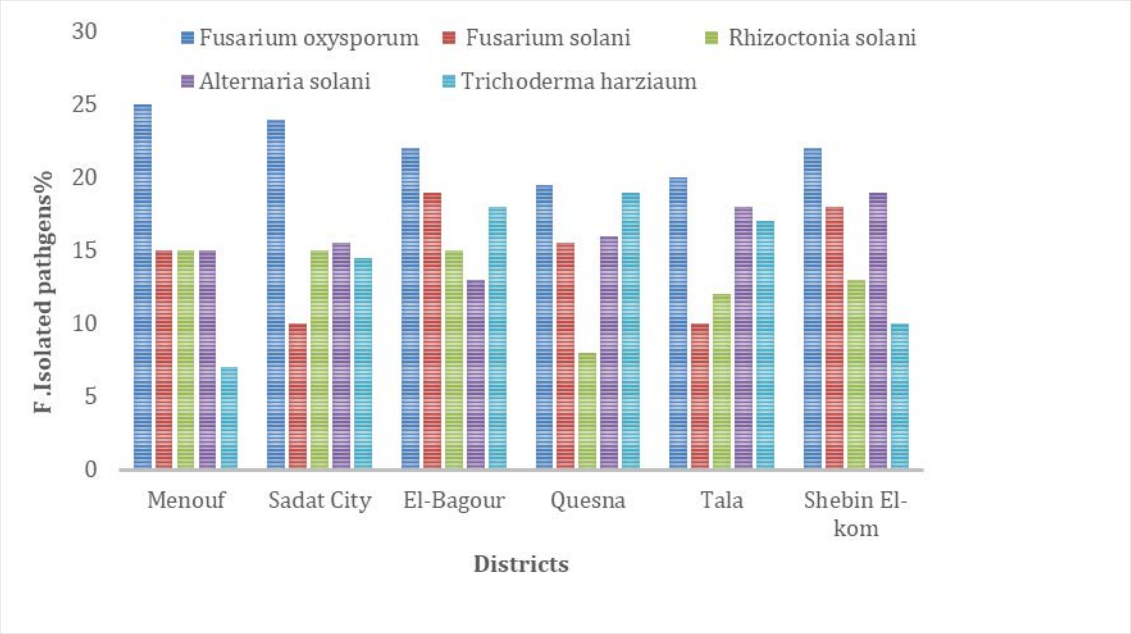

Fig. (1): Frequency of the isolated pathogens and biocontrol agents from different localities of Menoufia governorate.

Table (4): Pathogenicity test of *Fusarium oxysporum* isolates obtained from different localities of Menoufia governorate.

Isolate NO.	Province	Collapsed plants	Survival plant
FOF1	Menouf	4.67	0.33
FOF2	Sadat City	3.67	1.33
FOF3	Quesna	3.67	1.33
FOF4	El-Bagour	2	3
FOF5	Tala	0.67	4.33
FOF6	Sheben El-kom	0.67	4.33

3. Laboratory experiments:

3.1. Effect of plant extracts on the growth of (FOF1) (mm) after 2,4,6 and 8 days and the Av. No. of spores estimated after 8 days of plating the fungus.

Results present in Table (5) and clear that the plant water extracts significantly reduced the growth of (FOF) at all concentrations in comparison with control which estimated after 2,4,6 and 8 days. The best efficiency was observed when the extracts of either clove or cinnamon was applied to the pathogen even at the lower concentration (5%). Increasing the concentration of any tested extract resulted more inhibition of the fungal growth. On the other hand, the average number of spores was significantly decreased in response to the application of any tested extract; significantly. As the concentration was increased, the efficiency was increased and vice versa.

3.2. Effect of plant essential oils on the growth of (FOF1) growth (mm) after 2,4,6 and 8 days and the Av. No. of spores estimated after 8 days of plating the fungus:

The application of plant essential oils to PDA medium reduced the growth of (FOF1) significantly when estimated after 2,4,6 and 8 days of inoculation; compared to the nontreated control ones (Table 6). Cinnamon oil showed the best effect in reducing the fungal growth even at the lower concentration (2.5%). It was noticed that increasing the concentration of any tested oil led to more reduction of the fungal growth and vice versa.

The average number of spores was also significantly decreased in response to the application of any tested essential oil and increasing the concentration showed more efficiency.

Table (5): Effect of plant extracts on the growth of (FOF1) (mm) after (2,4,6 and 8 days) and the Av. No. of spores estimated after 8 days of plating the fungus.

Plant extract	Conc. (%)	Av. growth (mm) after (days)				Av. No. of spores /ml (×4000)
		2	4	6	8	
Clove	5	0.0^{g}	0.0^{g}	0.0^{f}	0.0^{g}	0
	10	0.0^{g}	0.0^{g}	0.0^{f}	0.0^{g}	0
	15	0.0^{g}	0.0^{g}	0.0^{f}	0.0^{g}	0
Cactus	5	13.6^{b}	26.6^{b}	57.3^{c}	79^{b}	27
	10	10.6^{c}	25.8^{c}	49.3^{d}	75.3^{c}	23.66
	15	8.6^{d}	21.3^{d}	42.3^{e}	69^{d}	17.33
Mint	5	14.5^{b}	32.6^{a}	67.8^{a}	90^{a}	28.66
	10	16.5^{a}	30^{b}	63.3^{b}	90^{a}	29.66
	15	14.6^{b}	29.6^{b}	63.6^{b}	90^{a}	30.33
Camphor	5	5.8^{e}	11.3^{e}	27^{f}	47.6^{e}	23.66
	10	5^{f}	9.6^{ef}	26.5^{f}	40^{ef}	19
	15	3^{fg}	8^{f}	21.1^{f}	32^{f}	11.66
Cinnamon	5	0.0^{g}	0.0^{g}	0.0^{f}	0.0^{g}	0
	10	0.0^{g}	0.0^{g}	0.0^{f}	0.0^{g}	0
	15	0.0^{g}	0.0^{g}	0.0^{f}	0.0^{g}	0
Control	_____	13.3^{b}	28.5^{b}	58.3^{c}	90^{a}	29.33
LSD5%		1.79	2.25	3.24	0.88	_____

Table (6): Effect of plant essential oils on the growth of (FOF1) growth (mm) after 2,4,6 and 8 days and the Av. No. of spores estimated after 8 days of plating the fungus.

OIL	Conc. (%)	Av. diameter of growth (mm) after (days)				Av. No. of spores /ml (x4000)
		2	4	6	8	
Clove	2.5	10.6^{fg}	28^{bc}	45.3^{def}	66.3^{d}	11.33
	5	11.6^{efg}	26.6^{bc}	38^{gh}	61.6^{e}	9
	10	9^{g}	24.33^{c}	43.3^{ef}	58^{e}	6.33
Garlic	2.5	17.3^{abc}	37.6^{a}	57^{ab}	74^{c}	18.33
	5	14.6^{bcde}	36.3^{a}	49.6^{cd}	69.3^{cd}	15.66
	10	0.0^{h}	18^{d}	28.3^{i}	44^{f}	11.33
Thyme	2.5	17.6^{ab}	36.6^{a}	55.6^{ab}	72.3^{c}	20.33
	5	15.6^{abcd}	28.6^{bc}	41.6^{fg}	70^{cd}	16
	10	14.3^{cde}	26.3^{bc}	36.6^{h}	47.6^{f}	12
Sesame	2.5	15^{bcd}	29.3^{b}	51.6^{bc}	79.3^{b}	21.33
	5	15.6^{abcd}	27.6^{bc}	46^{de}	76^{b}	20.33
	10	13.3^{def}	26.3^{bc}	46^{de}	73^{c}	18
Cinnamon	2.5	0.0^{h}	0.0^{e}	0.0^{j}	0.0^{g}	0
	5	0.0^{h}	0.0^{e}	0.0^{j}	0.0^{g}	0
	10	0.0^{h}	0.0^{e}	0.0^{j}	0.0^{g}	0
Control		18.3^{a}	39.3^{a}	58.3^{a}	90^{a}	27.66
LSD 5%		2.89	3.82	4.23	4.26	_____

4. Green-house experiments:

4.1. Cultivar susceptibility:

The susceptibility of three faba bean cultivars i.e., Australian, Balady and English to the infection with either *Fusarium oxysporum* isolates (FOF1, FOF2 and, FOF3) was studied. Results present in Fig. (2) clear that the local faba bean cv. Balady was the least susceptible one for the three *F. oxysporum* tested isolates. In the meantime, the imported Australian cv. showed that the most susceptibility to the tested isolates. English cv. was in between in this request Generally; the average number of collapsed plants was increased by increasing the time of estimation; 2,4 and 6 weeks after seeding in the artificially infested soil. The same observations were detected when estimated as growth parameters, severity of infection and spore population in the soil Fig. (2). Faba bean Balady cv. showed the highest plants and the least severity of infection which estimated as the average length of the xylem brown tissues Fig. (3); six weeks after planting. The average number of the pathogen spores into the soil was the highest with Australian cultivar in comparison with the other two tested cultivars.

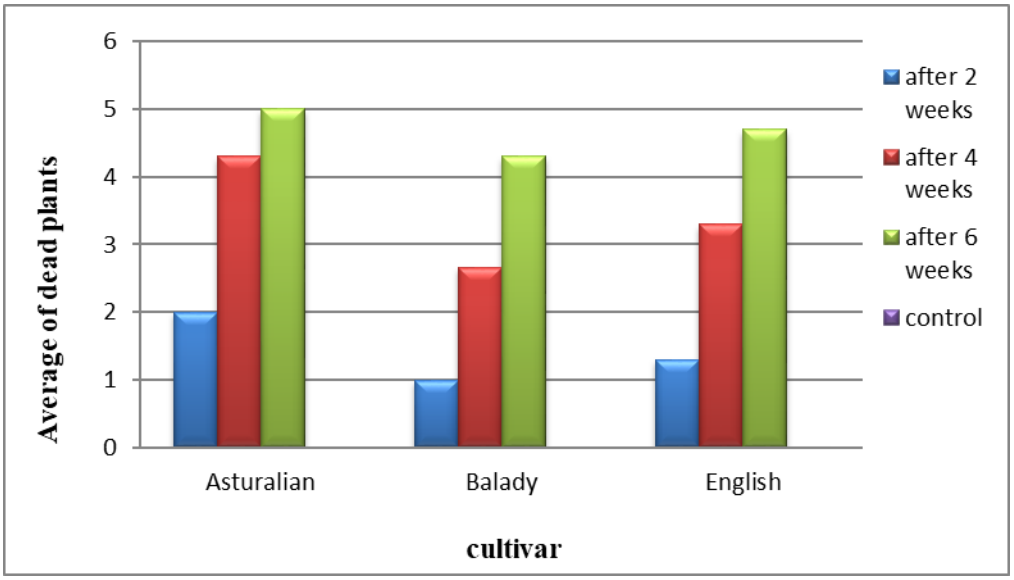

Fig. (2): Faba bean cultivar susceptibility to FOF1, FOF2 and, FOF3 isolates of *F.oxysporum* estimated as the average of dead plant 2-6 weeks old.

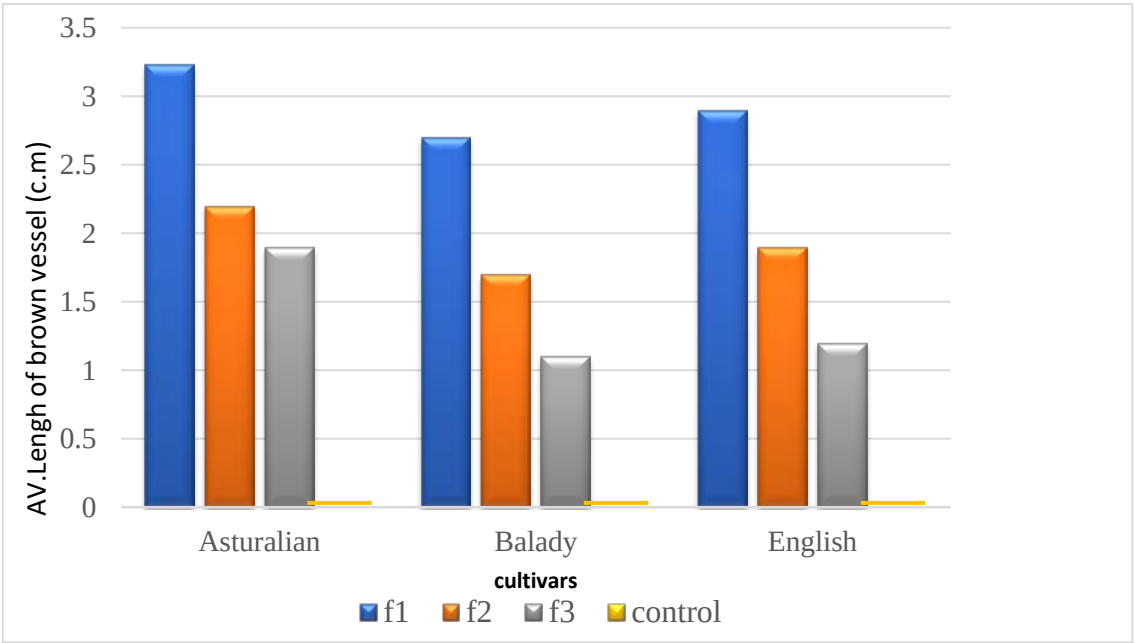

Fig. (3): Effect of (FOF) isolates (1, 2 and, 3) on faba bean cultivars growth, vesicles browning and spore population in the soil.

4.2. Control studies:

4.2.1. Effect of plant water extracts on pre- and post-emergence damping-off:

Results present in Table (7) and Fig (4) clear that clove and cinnamon were the best tested plant extracts for the disease control, where no pre-and/or post-emergence damping- off were happened at all tested concentrations (5, 10 and 15% (However camphor (15% (gave the same results. The least effective tested plant extract was mint followed by cactus.

Table (7): Effect of plant extracts on plant height, severity of infection with (FOF1) fungus and the Av. NO. spores in the soil.

Plant extract	Conc. (%)	Av. plant height (cm)	Av. Length of brown vesicles (mm)	Av. No. of spores/m (x4000)	R %*
Clove	5	40^{a}	0.0^{j}	0.0^{i}	-100
	10	41.3^{a}	0.0^{g}	0.0^{i}	-100
	15	42.3^{a}	0.0^{g}	0.0^{i}	-100
Cactus	5	28.6^{c}	10.26^{e}	30.3^{d}	-13
	10	28.3^{c}	8.33^{f}	23.3^{e}	-33
	15	43.6^{b}	7.4^{f}	17^{g}	-51
Mint	5	17.2^{e}	32^{a}	51.6^{a}	47
	10	19.3^{e}	30^{b}	50^{ab}	43
	15	17.6^{e}	29^{b}	48.3^{b}	38
Camphor	5	33.6^{b}	17.66^{c}	19.6^{f}	-44
	10	35.43^{b}	12.66^{d}	16.3^{g}	-53.5
	15	40.33^{a}	10.66^{e}	0.0^{i}	-100
Cinnamon	5	40^{a}	0.0^{g}	0.0^{i}	-100
	10	41.66^{a}	0.0^{g}	0.0^{i}	-100
	15	42^{a}	0.0^{g}	0.0^{i}	-100
control (-)		41.3^{a}	0.0^{g}	0.0^{i}	-100
control (+)		21.6^{d}	30^{b}	35^{c}	__
LSD 5%		2.85	1.19	2.89	

R*: Response of spores NO : + =Increment - = Reduction

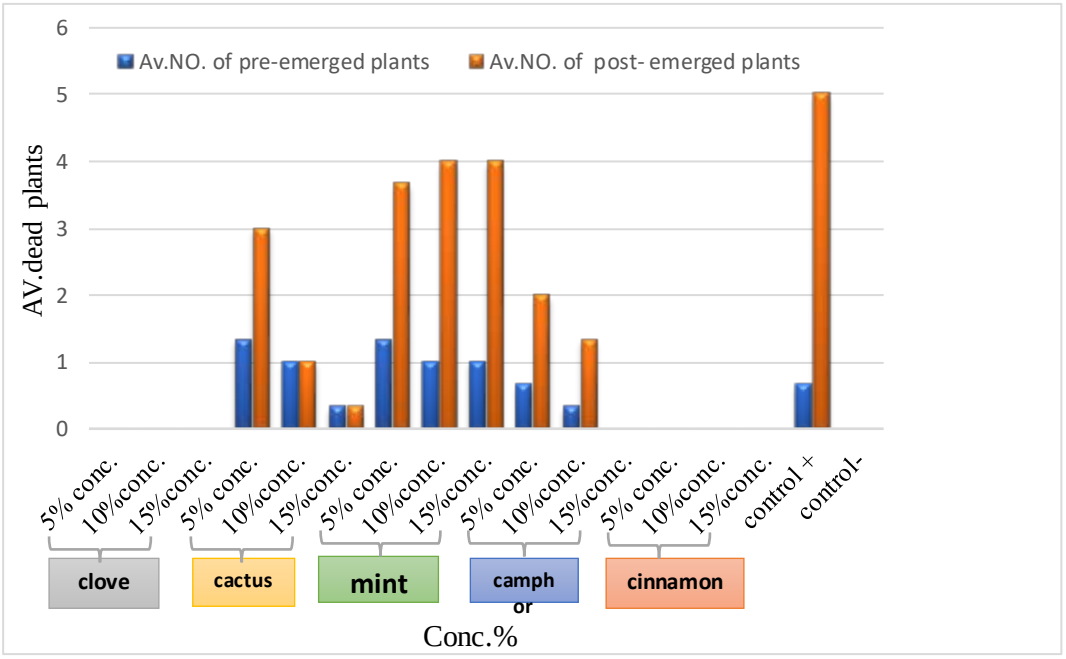

Fig. (4): Effect of plant water extracts on the pre- and post- emergence damping –off caused by *Fusarium oxysporum* isolate (FOF1) on Australian faba bean cultivar.

4.2.2. Effect of plant extracts on plant growth, severity of average No. of spores in the soil:

Results shown in Table (7) indicate that clove and cinnamon plant extracts gave the best action for controlling will disease of faba bean. Plant height was the best when either extract was applied at tested concentration. In the meantime, root vesicles were clear of browning both cases. Also, the average number of spores/ml in the soil was nil and 100% reduction was achieved! Application of mint plant extract gave opposite results; where plant height was less than control, the average length of brown vesicles was higher than control and the average number of Fusarium spores was increased by 47, 43 and 38%. in comparison with control (+) treatment. Such results clear that mint plant water extract encourage of the pathogen and increase its aggressiveness.

4.2.3. Effect of plant essential oils on pre-and post-emergence damping-off caused by (FOF):

Results present in Table (8) clear that cinnamon plant essential oil had the best efficiency in reducing pre- and post- emergence damping-off of Australian cv. Clove and garlic essential oils came in the second rank, where they were highly effective at 10% concentration. Sesame essential oil showed the least efficiency in reducing the disease.

Table (8): Effect of plant essential oils on the pre- and post- emergence damping –off caused by isolate (FOF1) on Australian faba bean cultivar.

Treatment	Conc. (%)	Av.NO. of pre-emerged plants	Av.NO. of post-emerged plants	Total NO. OF dead plants	R*%
Clove	2.5	0.66^{bcd}	0.66^{def}	1.32	38
	5	0.0^{d}	0.33^{ef}	0.33	66
	10	0.0^{d}	0.0^{f}	0	0
Garlic	2.5	0.66^{bcd}	0.66^{def}	1.32	38
	5	0.33^{cd}	0.33^{ef}	0.66	1.312
	10	0.0^{d}	0.0^{f}	0	0
Thyme	2.5	1^{bc}	1^{de}	2	40
	5	0.33^{cd}	0.66^{def}	0.99	20
	10	0.0^{d}	0.33^{ef}	0.33	66
Sesseme	2.5	1.6	3.4^{b}	5	100
	5	1.33	3.5^{b}	4.83	96.6
	10	1^{bc}	1.3^{d}	2.33	46.6
Cinnamon	2.5	0.0^{d}	0.0^{f}	0	0
	5	0.0^{d}	0.0^{f}	0	0
	10	0.0^{d}	0.0^{f}	0	0
Control (+)		0.66^{bcd}	5	5	100
Control (-)		0.0^{d}	0.0^{f}	0	____
LSD 5%		0.61	0.66	_____	____

R% : Reaction % compared to control (+)

4.2.4. Effect of plant essential oils on plant growth, severity of infection and the average No. of spores in the soil:

Results present in Table (9) clear that all growth of faba bean plant in comparison with control (+) treatment. Cinnamon tested plant essential oils significantly improved the gave the best effect Allowed by chore oil. Such treatments. Also were effective in reducing the average length of the brown vesicles, significantly. Spore population in the soil was highly oils. It could be noticed that cinnamon essential oil at all tested concentrations (2.5, 5 and 10 %) and clove oil (10%) suppressed the growth of the fungus in the soil, where no Fusarium spore were present after 45 days. In the meantime, sesame essential oil showed the least efficiency in reducing spore population in the soil.

Table (9): Role of plant essential oils on plant (45 days after seeding), severity of infection with isolate (FOF1) and the Av. No. of spores in soil.

Treatment	Conc. (%)	Av. plant height* (cm)	Av. Length of brown vesicles (mm)	Av. No. of spores/ml (x4000)	R*%
Clove	2.5	40^{cd}	6.33^{h}	14^{de}	46.6
	5	41.3^{bcd}	2.33^{i}	6.33^{g}	21.1
	10	43.6^{a}	0.0^{j}	0.0^{h}	0
Garlic	2.5	38.6^{efg}	10.26^{f}	16.3^{d}	54.3
	5	39.6^{def}	8.33^{gh}	11.6^{ef}	38.7
	10	41.3^{bcd}	7.4^{g}	0.0^{h}	0
Thyme	2.5	34^{h}	17.66^{d}	22.3^{b}	74.3
	5	38^{fg}	12.66^{e}	17.6^{cd}	58.7
	10	37.1^{g}	10.66^{f}	6.6^{g}	22
Sesseme	2.5	27.6^{j}	22^{b}	28.3^{a}	94.3
	5	31.1^{i}	20^{c}	21^{c}	70
	10	31.6^{i}	19.16^{c}	14^{de}	46.6
Cinnamon	2.5	40^{cde}	0.0^{j}	0.0^{h}	0
	5	41.8^{bc}	0.0^{j}	0.0^{h}	0
	10	42.6^{ab}	0.0^{j}	0.0^{h}	0
control (-)		41.3^{bcd}	0.0^{j}	0.0^{h}	0
control (+)		21.6^{k}	30^{a}	30^{a}	___
LSD%		1.71	1.28	3.49	___

R* =Reduction of spores in the soil% after estimated 45 days seeding after compared to control (+).

Discussion

Fusarium oxysporum f. sp. *fabae* was also reported to be the causal organism of faba bear wilt disease by Lvjia Xing *et al*., (2020), Li Yu *et al*., (2020) and Jinxing *et al*., (2021).

However, variation in isolates percentages obtained from different locations could be due to the environmental condition or the cultivated faba bean cultivar and for both together.

However, susceptibility and for resistance of faba been cultivars to wilt disease were also studied by Zaitoun and Ter Borg (1994) and Mahmmoud and El-Fatah (2020).

Several formulated botanical extracts were shown to effectively reduce soil populations of *F. oxysporum* and increase symptomless plant stand in controlled experiments. The suppression of wilt development in the greenhouse corresponds with the ability of these extracts to reduce populations of Fusarium in soil.

Clove and plant cinnamon water extracts were the best ones for controlling *Fusarium oxysporum* and other soil-borne fangi by Huda ahmed and Bayounis (2019) and Kamei, *et al.*, (2022).

We do not decide, at this time, that these products by themselves in their current formulations can replace fungicides or other fumigants and pesticides on a one-for-one basis in all situations. However, these extract formulations may develop into components of different management strategies and would depend on the cropping system. However, the observed reduction in the pathogen population indicates that these extracts could have important roles in biologically based management strategies for controlling Fusarium wilt diseases.

Preliminary data suggest that some of these extracts and essential oils are capable of pathogen growth inhibition in-vitro when tested so that only volatiles interact with the pathogen, while others only inhibit the pathogen when in direct contact. Information of this type is important as one tries to develop a delivery system that utilizes the extracts physical and chemical properties (Bowers, *et al*., 2000). Hiroko *et al*., 2016 analysis the composition of clove extract by GC-mass Chromatography, they suggest the effect of clove, attributed to the Phenolic compounds such as eugenol, eugenol acetate, and gallic acid which are recognized as a major antioxidant in cloves composition. We suggest that These compounds play an important role with the antioxidative activity in inhabiting the fungal growth.

The composition analysis of cinnamon leaf extract by GC-mass Chromatography was tested by (Anak *et al*., 2019), they conclude that cinnamon composition consists of 16 compounds which are dominated by 3 compounds namely 1,2-Benzenedicarboxylic acid, mono (2-ethyl (29%), 2H-I-Benzopyran-2-one (CAS) Coumarin (11.9%) and 2, 6- dimethyl-6-nitro-2-hepten-4-one (11.5%), these compounds play an important role of the antifungal activity naturally.

The antagonistic and growth promotion potential of Trichoderma against *Fusarium oxysporum* isolates were also reported by Pandy Ghanshyan *et al*., (2021).

Ivanovic *et al*., (1987) observed that *Fusarium oxysporium* f. sp. *fabae* was also pathogenic to soybean, Phaseolus, vulgaris and pea. Host rang experiments of (FOF) were also conducted by Hassanin *et al*., (1997) and Infantion *et al*., (2006).

The antifungal activity of clove and cinnamon plant extracts against *Fusarium oxysporum* was also observed by Kishore and Srikarni (2008) who mentioned that clove extract and clove oil were the best for reducing *Fusarium oxysporum* spore population in the soil and improved plant growth.

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تأثير بعض المستخلصات النباتية والزيوت النباتية على مرض الذبول الفيوزاريومي في الفول

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الملخص العربى

فطر الفيوزاريوم *Fusarium oxysprium* f.sp. *fabae* المسبب المرضي لمرض الذبول في الفول تم عزله من عدة مراكز (منوف -السادات -الباجور-قويسنا-تلا- شبين الكوم) داخل محافظة المنوفية. تجربة اختبار العدوي أثبتت أن أكثر العازلات ضراوة هو جنس الفيوزاريوم الذي تم عزلة من مركز منوف. تم تقييم ثلاث أصناف من الفول (الأسترالي-البلدي والإنجليزي) لبيان أكثر الأصناف قابلية للإصابة مع أكثر الفطريات ضراوة (عزلة منوف) تحت ظروف الصوبة. في تلك الدراسة تم استخدام خمس مستخلصات نباتية (النعناع – القرنفل – القرفة – الصبار والكافور) بتركيزات (٥ -١٠ و١٥ %) وكذلك تم استخدام خمسة زيوت نباتية (القرنفل -القرفة -السمسم - الثوم والزعتر) بتركيزات (٢,٥ -٥ و١٠٪) لبيان مدي تأثيرهم على تثبيط النمو الفطري وعلى تجرثمه تحت ظروف المعمل والصوبة. تحت ظروف المعمل تم اختبار كل المستخلصات النباتية والزيوت النباتية منفصلة حيث أدت الي تقليل متوسط خط النمو وقابليته على التجرثم وكانت أكثر المستخلصات فاعلية هما مستخلصي القرفة والقرنفل بينما كان زيت القرفة هو الأكثر تأثيرا عند التركيزات القليلة. تحت ظروف الصوبة وإجراء عمليات العدوي الصناعية للفطر المسبب ادي استخدام المستخلصات النباتية والزيوت النباتية إلى تقليل شدة المرض وأيضا قدرته على التجرثم معنويا.