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PATHOGENICITY OF *Fusarium solani* AND *Ilyoncteria macrodidyma* ON OLIVE SEEDLINGS ROOTS AND THE EFFICACY OF SOIL MICROBIAL COMMUNITY AGAINST INFECTION

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ABSTRACT

A total of six olive fields were inspected randomly in three locations (Sartank, Sharia and Qasrook) at Duhok province. The highest severity (40%) and incidence (83%) of olive tree wilting and dieback were recorded in Sharia while the lowest severity was in Sartank (35%). The predominant fungi in this study were *Fusarium solani* and *Ilyoncteria macrodidyma*. The occurrence percentage of isolated fungi varied between locations. *F.solani* was the most frequent (53.3 %) on Qasrok, while *I. macrodidyma* was recorded at low frequencies (16.6%). *F. solani* was isolated by 29.4% and 27.2% in Sharia and Sartank respectively. Disease severity of *F. solani* in aerial part of olive seedlings during May, July, and September was 22.66%, 40.33% and 67% respectively significant differences, while was 24.67%, 31%, and 45.33by *I. macrodidyma* % respectively. Pathogenicity after five months showed high root infection by *F. solani* (100%) incidence and (68.20) severities. The reduction in wet and dry weight of the green part was 25.75% and 36.64% and in roots system was 59.29% and 50.99% respectively. *I macrodidyma* showed (100 %) disease incidence and (46.70%) severity in root system. The reduction in wet and dry weight of the green part was 18.23%, 18.80%, while in olive root was 34.66%, 29.32% respectively.

***Trichoderma harzianum* rapidly stopped *F. solani* mycelia growth, and showed (64%) inhibition, while *Gliocladium sp* inhibited the radial growth of *F. solani* to (31%) and *Aspergillus niger*, (22%) inhibition respectively. High in vitro inhibition of *I. macrodidyma* was observed by *T. harzianum* (77%). The results of dual culture method revealed that *T. harzianum* completely overgrew the pathogen *I. macrodidyma* and was placed in class1 according to Bells scale; whereas *Penicillium sp*. parasitized the tested pathogen up to two-thirds of the medium and were placed in class 2. The antagonism against *F. solani* showed the overgrew of *T. harzianum* up to two thirds of the medium and were placed in class 2, while the pathogen colonized at least two-thirds of the medium and overgrew the *Gliocladium sp* and *Aspergillus niger* colonies and were placed in class 4. *Penicillium sp*. colonized one-half of the medium and was placed in class 3.**

KEYWORDS: *Fusarium solani , Ilyoncteria macrodidyma, Cylindrocarpon sp,* Olive , *Trichoderma harzianum, Gliocladium sp, Aspergillus sp, Penicillium sp*., Pathogenicity

INTRODUCTION

F. solani is a soil-borne fungus that affects a wide range of crops, vegetables, and trees, causing a variety of symptoms such as seed rotting, mortality, and damping-off seedlings, as well as dry root rot (Kunta et al., 2015 and Al-Karboli and Kuthair, 2016). According to Trabelsi et al., (2017), the most significant and serious disease affecting olive trees is dieback and wilting symptoms produced by complicated soil-borne fungi, *Fusarium* is one of the most important phytopathogenic genera linked to olive tree dieback. According to the pathogenicity test, *Fusarium spp*. could be a major cause of olive tree dieback in Tunisia. When examining the pathogenicity of *Fusarium* species obtained from olive trees and identifying them based on morphological and genetic characteristics, the incidence of the disease has increased steadily over the years and appears to be associated with changes in agricultural practices and a loss in plant growth.

Cylindrocarpon (*Ilyonecteria*)-like species associated with black foot disease are symptoms

of grapevine root rot and roots in other perennial crops such as almond, cherry, kiwi, olive, peach, pistachio, and walnut. Black root rot, stunted growth, yellow leaves, root sunken, necrotic lesions, and in most cases reaching the base and crown of the trees are all the symptoms of black foot disease on trees. Symptoms of leaf wilting and chlorosis, brown to black wood discoloration in cross-sections of the stems, and necrotic lesions in the roots were found on three- to ten-year old olive trees (Úrbez-Torres et al., 2012).

Researchers are interested in the use of microorganisms as a source of bioactive substances (Bertrand et al., 2014). Due to the great biological activity potential of microbe's large-scale fermentation processes can reduce the costs of microbe-derived compounds. Antagonists are among plant-associated microbes with great potential to provide bioactive metabolites that can inhibit the growth of pathogens, in part or total (Van Eeden and Korsten, 2013).

Trichoderma spp. is considered one of the most common biological control agents, and it has undergone extensive testing and studies around the world. Chitinase and glucanase, with several other enzymes that degrade the cell wall of fungi have important role in the antagonistic action of *Trichoderma* effect on fungal pathogens (Kucuk and Kivanc, 2008). The biological control agents *T. harzianum* and *Gliocladium* have proven relatively high efficacy against *Fusarium solani* (Butt et al., 2001). Suppression of Fusarium species by *Gliocladium catenulatum* have been reported by Syama et al. (2008) indicating the high suppression effect on Fusarium species. *G. catenulatum*, for example destroys the mitochondrial cells of *Sclerotinia sclerotiorum* and *Fusarium spp* by direct interaction without penetration, causing the affected cells to collapse break down, or disintegrate, according to Huang, (1978). In A different isolate, was found to coil around and penetrate the hyphae of *Rhizoctonia solani*, resulting in granular and disintegrated cells (Turhan, 1990).

Jha and Jalali, (2006) tested the biological control ability of pea roots fungi isolates against *F. solani* in vitro (dual culture method) and diseased soil conditions (pot). In a dual culture experiment, *T. viride* demonstrated the strongest antagonistic action against *Fusarium solani* f.sp. *pisi*. The biological regulation of pea root rot under pathogen diseased soil (pot) conditions, investigated separately and in conjunction with the two highly effective antagonists, *T. viride* and *Aspergillus niger*. *T. viride*, *T. harzianum*, *T. aureoviride*, *T. koningii*, *T. pseudokoningii*, *A. fumigates*, *A. Glacus*, and *A. oryzae* were shown to be involved in control wilt pathogen (*F. solani*) Arshad, (2008). In dual culture techniques, Gupta and Mishra, (2009) observed that *A. niger* was superior in inhibiting the growth of *F. solani*. The use of non-pathogenic as, *Penicillium oxalicum* and *F. oxysporum* in numerous experiments reported a significant reduction in the incidence and severity of *Fusarium* tomato wilt disease (Larena et al., 2003). Van Jaarsveld et al., (2021) demonstrated that *Trichoderma* can inhibit the growth of black foot disease (BFD) pathogens in vitro a competitive growth study and revealed that all *Trichoderma* isolates exert some form of antagonism against black foot disease pathogens, and *D. macrodidyma* was found to be the most sensitive pathogen. The aims of this study is Determination the severity and incidence of olive root rot and wilt disease in three major olive producing area of Duhok province; Isolation and pathogenicity of *Fusarium solani*, and *Ilyonectria macrodidyma* and knowing the fungal community in the soil of olive orchards to determine their association with this disease

MATERIALS AND METHODS

1. Field Survey and fungal Isolation

Disease survey was carried out in three major olive production areas in (Sartank, Sharia and Qasrook) in Duhok province, by using square unit's method. Percentages of diseased trees, showing symptoms of root rot and/or wilt diseases were recorded in July, Fields were randomly selected in each of the previously mentioned area, three samples were taken from each unit, the total number of samples from each field was fifteen. Disease severity (DS) was calculated on naturally affected trees by using the following formula:

Disease Severity (%) = $\sum$ (n×v)/ N×V ×100. Where: n= is the number of diseased trees per category. v= Category number. N= Total number of the trees. V= Maximum disease severity rate. The disease severity was assessed on trees that showed typical symptoms of chlorosis and wilt based on the foliar symptoms, dull green, internally wrapped or necrotic leaves and defoliated twigs were included (El-Morsi et al.,

2009). A scale of 0-4 on the basis of the rating were used, 0= trees healthy. 1=0 to 25% (Milled symptoms). 2=<25 to 50% (Intermediate symptoms). 3=<50 to 80% (Severe symptoms). 4=<80% Diseased foliage (tree is death).

Olive roots samples were collected from (12-15) years old trees from Sharia, Sartank and Qasrok sub- district – Duhok province which is considered as the main site for olive production in Kurdistan region- Iraq. The collected roots were washed under running tap water for (30) minutes. They were cut into small parts of (1 cm) and surface is sterilized by sodium hypochlorite (2%) for (3) minutes. They were rinsed in sterile distilled water for three minutes and dried between two filter papers. Five root pieces were put on petri dishes containing Potato Dextrose Agar (20 ml) and chloramphenicol (250mg/L) to prevent bacterial growth, and incubated at 25 ℃ for 7 days. The frequency of each fungus was determined as follows Frequency % = Colony No. of isolated fungus/plate (x) / Total No. of colonies/plate × 100. Fungal growth was purified by single spore technique and identified based on morphological and microscopically features (Booth, 1966; Úrbez-Torres, et al. 2012 and Lawrence, et al. 2019). Purified isolates were kept on PDA slants and stored in the refrigerator at 5 °C.

2. **Pathogenicity Test:**

The inoculum of *I. macrodidyma* and *F. solani* were prepared from two weeks old cultures grown on PDA at 25±1°C. Inoculations were carried out by dipping each individual's bare root system of nine seedlings of two years old into the conidial suspension (4×10^6 conidia/ml) for half an hour. In non-inoculated treatment, the roots of control plants were dipped in distilled sterile water. Plants were individually transplanted into sterile plastic pots (20 cm) containing sterile soil (1:2, Sandy loam soil: Peatmous) under greenhouses condition ±28 °C. The plants were arranged in RCBD design with three replications. Data were recorded during May, July and September 2019 depending on the percentage of foliage with yellowing or necrosis, the severity of the aerial symptoms was assessed for each plant on a scale from 0 to 4. 0 = 0 trees healthy, $1 = 1 - 33\%$ (Milled symptoms), $2 = 34 - 66\%$ (Intermediate symptoms), $3 = 67 - 99\%$ (Severe symptoms), $4 =$ Dead plant. The same 0–4 scale was used to measure root rot and discoloration during September (after five months from inoculation) (Sánchez-Hernández et al., 1998). The reduction in the dry and wet weight of root and green growth was also recorded. Data analysis was carried out by SAS and the means were compared using the Duncan's multi-range test (SAS 2003).

3. Fungal dynamic associated with olive root and rhizosphere.

3.1. Collection of soil samples and assessment of soil fungi communities.

Soil samples were randomly collected from olive surveying sites in Duhok governorate. About 500 grams of soil samples were collected from 0-20 cm depth by trowel after disposal of litter or weed plants. Soil samples were placed in plastic bags marked with the labels, and then transferred to the laboratories of the Plant Protection Department, which stored at 4°C for further isolation. Fungal isolation occurred within 2 days of sample collection. Prior to use, samples were mixed well and passed through a fine mesh screen to break down clumps of earth and separate litter debris. Soil dilution plate method was used to analyze air-dried samples for 4 weeks at room temperature (Waksman, 1922).

Plate of Soil dilution: Soil samples for each core were independently blended after being thoroughly smashed. 10g of the soil blend in distilled in 90ml of sterile distilled water (w: v) at that point series of dilutions 10^{-1}, 10^{-2} and 10^{-3} were prepared. Approximately 0.2 ml of volume for final dilution was spread on 2% dextrose agar (PDA), and then incubated at 25 ± 1°C. Data for single spore isolation techniques were used to purify all isolates and store them in PDA medium at $4°C$ for further analysis, using light microscopic examinations and standard diagnostic keys to check colony features, mycelial growth, and the presence of conidia (Barnett & Hunter, 1986).

3.2. In vitro efficacy of antagonistic fungi:

Gliocladium sp., *A. niger*, *Penicillium digitatum*, and *T. harzianum* were isolated and tested for antagonism against pathogenic fungi *F. solani* and *I. macrodidyma* using the dual culture technique (Rama et al., 2000) in PDA petri dishes as described by Morton & Strouble, (1955). Pathogenic mycelial plugs of 7 mm in diameter were obtained from the edge of colony of 7 days from each isolate grown in the PDA and then placed on the periphery of culture plates. The fungal plug was placed at 4 cm from the pathogen plug and incubated for a week at 25

± 2 °C in the dark. Three plates were inoculated with pathogen plug. Each pathogen / antagonist combination in the randomized complete block design was repeated three times. The radial growth of the pathogen in the double culture and the control plates were measured at a temperature of 28 ± 1 ° C as described by Vincent & Budge, (1990) after the pathogen growth completely covered the petri dish in control treatment. The formula proposed by Vincent, (1947) was used to calculate the inhibition rate of mycelial growth of test organisms over control as follows:

$I(\%) = \frac{C-T}{C} \times 100$; Where, I = Inhibition rate, C = controlled Growth, T= Growth in treatment.

The degree of antagonisms between each bio agent and pathogen were also tested in a dual culture method and recorded on a scale of 1-5 as suggested by Bell et al., (1982). First, the antagonist was completely overgrew the pathogen and covered the medium surface , Second, the antagonist overgrew at least 2/3 of the medium surface, Third, both the antagonist and the pathogen expanded on one half of the medium surface (greater than 1/3 and smaller than 2/3) and none of the organisms appeared to control each other. Forth: The pathogen colonized at least 2/3 of the medium surface and appeared with the stand overtaking. Fifth, the growth of the pathogen against the antagonist was successful and the entire surface of the medium was colonized.

RSULTS AND DISCUSSION

1. Field Survey

Disease severity was assessed as trees exhibited symptoms associated with olive tree branch wilting and dieback or tree death of root rot and wilts disease depending on foliar symptoms, including dull green, internally rolled or necrotic leaves and defoliated twigs, this observation was also noticed by El. Morsi, et al. (2009). The highest disease severity (40%) and incidence (83%) were recorded in Sharia. Meanwhile, the lowest founded in Sartank (35%) disease severity and (83%) disease incidence (Table 1).

Table (1): Disease severity and incidence on olive trees in Duhok province, during July and August 2019

Location	Disease severity	Disease incidence
Qasrok	40%	74%
Sharia	40%	83%
Sartank	35%	83%

The dieback and wilting symptoms induced by complex soil-borne fungi have caused significant economic losses in olive orchards in other areas (Úrbez-Torres et al., 2012; Chliyeh et al., 2014 and Gharbi et al., 2015). *Fusarium* and *Ilyoncteria* species are among the fungus that can cause this disease in olive trees. The presence of a wide variety of fungus isolated from the affected plant tissues was discovered during the diagnosis of dieback and decline in young olive trees (Triki et al., 2009 and Chliyeh et al., 2014).

At each of the three places inspected, the disease incidence and severity were different. Warm and dry environment in these regions, as well as long-term cultivation without adequate sanitation and preventative treatment control measures, may be attributed for the highest rates of disease incidence and severity. It seems inoculum density and distribution in the soil, the existence of mechanical or natural pathogen dispersal mechanisms within an orchard, the virulence of the isolates prevailing in the soil, cultivar resistance, the number of years that other herbaceous hosts were grown in the plot before olive was planted on it, and orchard age appear to all play a role. Such observations are in agreement with those reported by Barrera et al., (2003); El- Morsi et al., (2009); and Yaseen and D'Onghia, (2012).

2. Isolation of the causal agents of olive root rot:

Symptomatic trees exhibited black root rot; Chlorotic leaves with black, sunken, necrosis on

the roots and stunted growth, sometimes touching the trees' base and crown. On three- to ten-year-old olive trees, Signs of wilting and chlorosis of the leaves discoloration of brown to black wood in cross-sections of the stems, and necrotic lesions in the roots have been found. The predominant fungus in this study was *F. solani* and *I. macrodidyma* isolated from olive from districts in Qasrok, Sartank and Sharia. *F.solani* and *I. macrodidyma* has been isolated as the most common fungal pathogen from roots crown and stem of olive in many field survey from different areas of the world (El-Morsi et al., 2009; Úrbez-Torres et al., 2012; Triki et al., 2014 ; Chliyeh et al., 2017; and Ben Amira et al., 2018) Results of isolation indicates that *F.oxysporum*, *F.solani*, *R.solani*, *Macrophomina spp*, *Cylindrocarpon sp*., *Acremonium sp* and *Chaetomium sp*. were the most frequently isolated fungi from roots of olive trees which showed typical symptoms of root rot and wilt disease complex collected from different locations in Qasrok, Sharia and Sartank. Frequency of the isolated fungi varied between locations. Generally, *Fusarium spp*. was the most common pathogens in these districts. *F. solani* was the most frequent (53.3%) on Qasrok, while *Ilyoncteria spp*. recorded at low frequencies (16.6%). *F. solani* was isolated by (29.4%) in Sharia and by (27.2%) in Sartank.

Depending on geographical location, weather, and cultural practices (Daami-Remadi, 2006) and physiology of the host crop (Tivoli et al., 1986 and Isaac et al., 2018), the severity and impact of these pathogens are determined; the dominant isolated fungus was *Fusarium spp*. may potentially play a role in the dieback syndrome as a fungal complex.

3. Pathogenicity of fungi associated with olive root rot:

Typical symptoms of *F.solani* on infected plants were observed five months after inoculation. Seedlings showed leaf curling, yellowing and browning. On the root and crown, there was necrosis and dark discoloration, as well as browning of the vascular tissue (Fig.1 A1, A2). The same symptoms were noticed by (Kunta et al., 2015; Trablesi, 2017). Olive seedlings inoculated by *I. macrodidyma* showed clear and brownish color discoloration. Some basal leaves died, and others wilted, exhibiting chlorosis, leaf edge necrosis, curling and die back. The crown and root vascular were necrotic, sunken with reduction in secondary roots (Fig.1 B1, B2)

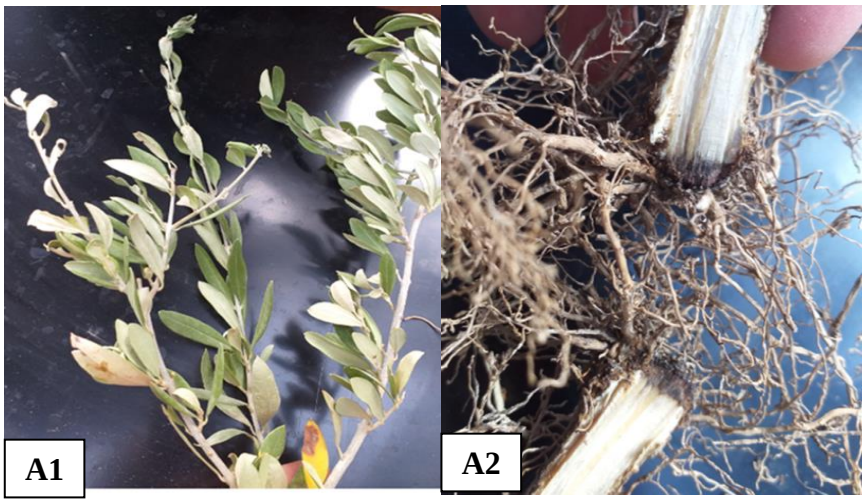

Fig (1): (A)Symptoms of *F. solani* A1- Seedlings leaf curling and yellowing. A2- Browning discoloration on the inoculated root (B) *I. macrodidyma* . B1- Chlorosis, leaf edge necrosis, curling and die back. B2-. Necrotic, sunken vascular with browning roots discoloration.

Disease severity of *F. solani* in aerial part of olive seedlings during May, July, and September was 22.66%, 40.33% and 67% respectively significant differences, while the disease severity of *I.macrodidyma* was 24.67%, 31%, and 45.33% respectively. Morphologically, isolates recovered from symptomatic plants were recognized as *F. solani* and *I. macrodidyma* with (100%) diseases incidence. No symptoms were shown by non-inoculated control plants (Fig 2).

At five months, Pathogenicity results showed of *F. solani* showed high disease incidence (100%) and severity (68.20) in root system. The reduction in wet and dry weight of the green part was 25.75 and 36.64 respectively (Fig 3). The root system of inoculated olive seedlings showed brownish discoloration and numerous decaying roots with high reduction in wet and dry weight by 59.29 and 50.99 respectively. Control plants had asymptomatic root systems.

Pathogenicity test of *I.macrodidyma* observed (100 %) disease incidence and (46.70%) diseases severity in root system. The reduction in wet and dry weight of the green part was 18.23%, 18.80% respectively, while olive root observed highly reduction by 34.66%, 29.32% (Fig 3).

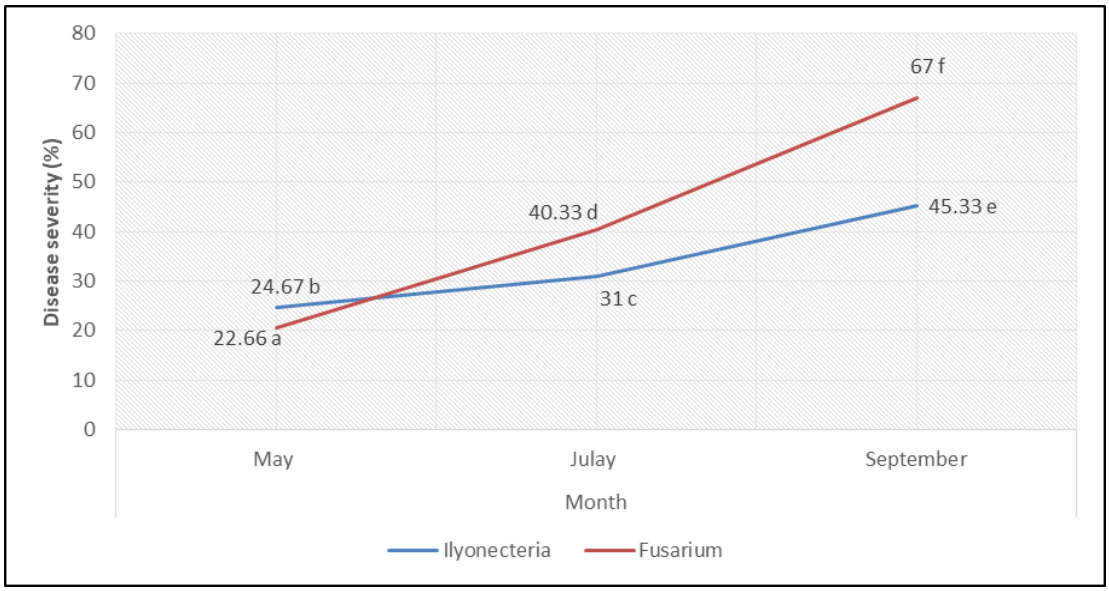

Fig. (2): Diseases severity of root rot wilt disease on olive transplants during May, July and September 2019.

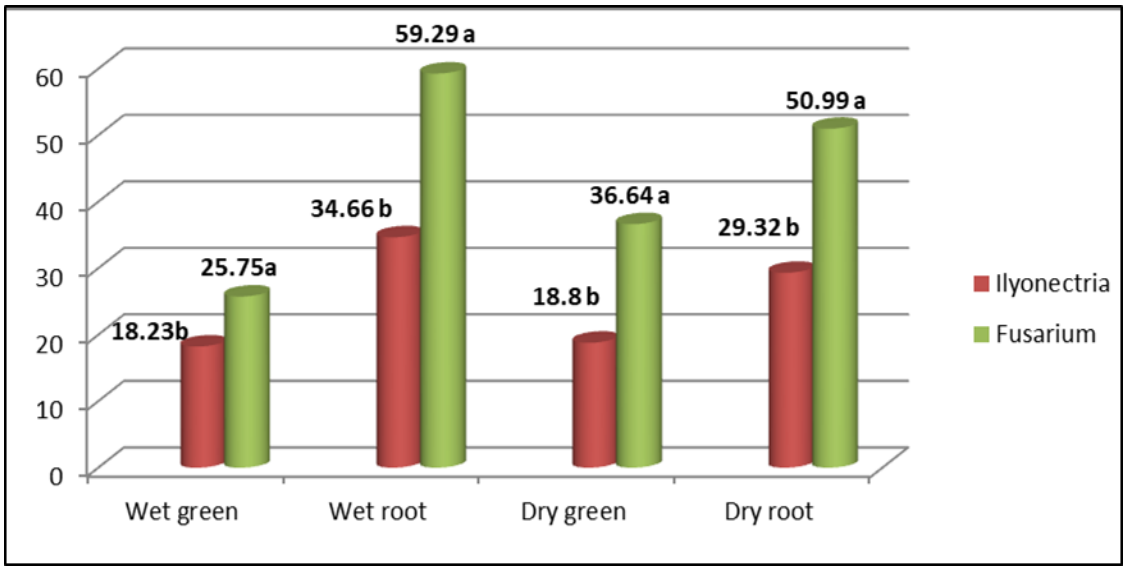

Fig. (3): Effect of disease on the reduction rate of wet and dry weight of root and green parts.

When the seedlings were inoculated with *F.solani*, they were smaller than the non-inoculated with a 19.63% reduction in plant length, whereas the inoculated seedlings with *I.macrodidyma* showed 21.46% reduction compared with non-inoculated (Fig. 4).

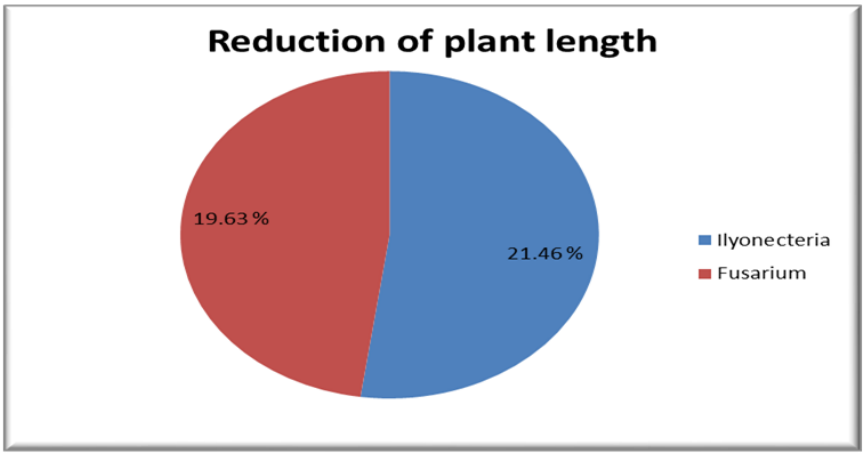

Fig. (4): Effect of disease on the reduction rate of plant length.

These results are in agreement with other studies (Teviotdale, 1994; Ghoneim et al., 1996; Sánchez-Hernández et al., 1998 & 2001 and Barreto et al., 2002). Generally, the results indicate clearly that *Fusarium spp*. were the most common pathogens followed by *I.macrodidyma*. Several factors may interact with incidence of diseases on olive trees (Martelli et al., 2002). The plant material and rooting conditions may affect the infection by certain fungal pathogens (Teviotdale, 1994). Latent infections may spread during rotting phase (Martelli et al., 2002). High humidity conditions accomplished by mist treatment may favor certain fungal pathogens. Farh et al., (2018) illustrated how attack *Ilyonectria sp* to root system that root-rot occurs due to compatible interactions between the root and the aggressive *Ilyonectria* species when the pathogen spore or mycelium adhere to the root surface, it quickly produces high number of hydrolytic enzymes such as cellulose and pectinase, enabling for quick invasion of the epidermal layer and rapid spread of the inoculum into the cortical and inner tissues. As the plant starts to respond to the invasion by releasing

phenolic compounds, the pathogen responds by created enzymes that can break down phenolic compounds such as polyphenol oxidases.

4. In vitro efficacy of antagonistic fungi:

T. harzianum rapidly stopped *F.solani* mycelia growth, and covered the *Fusarium* colony (Fig 5). *Fusarium* hyphal growth definitively ceased and inhibited at 64%, *Trichoderma* colony able to encroach into the inhibition zone and overlap the *Fusarium* colony. Other studies were also observed the *Trichoderma* activity against *Fusarium spp* (Eshetu et al., 2015; Amira et al., 2017). *Trichoderma* colonized a large area of the culture medium due to its faster mycelial growth than *F. solani*. This ability to grow rapidly gives the antagonist a notable advantage in competition for space, nutrients and dominance over its prey host. all being equally important and mutually inclusive phenomena (Altomare et al., 1999; Benitez et al., 2004).

Data presented in (Fig.5) indicated that the bioagent *Gliocladium sp* inhibited the radial growth of *F. solani* to 31% and caused reduction in the colony diameter of the pathogen. This found agree with Ben Salem et al., (2011). *Aspergillus sp.* showed 22% inhibition against the growth of *F. solani* .The inhibition effect of *F. solani* by *Aspergillus spp*. has been also reported by others (Getha et al., 2005; Gachomo and Kotchoni, 2008). The percentage reduction in colony growth of *F.solani* by *Penicillium sp.* presented in (Fig.5) revealed an increase in the bioagent colony with growth reduction of *F. solani* at 42%. The inhibition effect of *F. solani* by bioagents have been widely exploited in the management of soil-borne diseases (Fahri and Murat, 2007; Jayaraj et al., 2007).

High in vitro inhibition of *I. macrodidyma* was observed by *T. harzianum* (77%). This observation agrees with van Jaarsveld et al., (2021). *Aspergillus sp., Gliocladium sp.* and *Penicillium sp* noted 40%, 44% and 63% inhibition percentage against the growth of *I. macrodidyma* respectively (Fig.5). In-vitro antagonistic of *T. harzianum* and *Gliocladium sp.* has been also proven against *F.solani* by Butt et al. (2001).

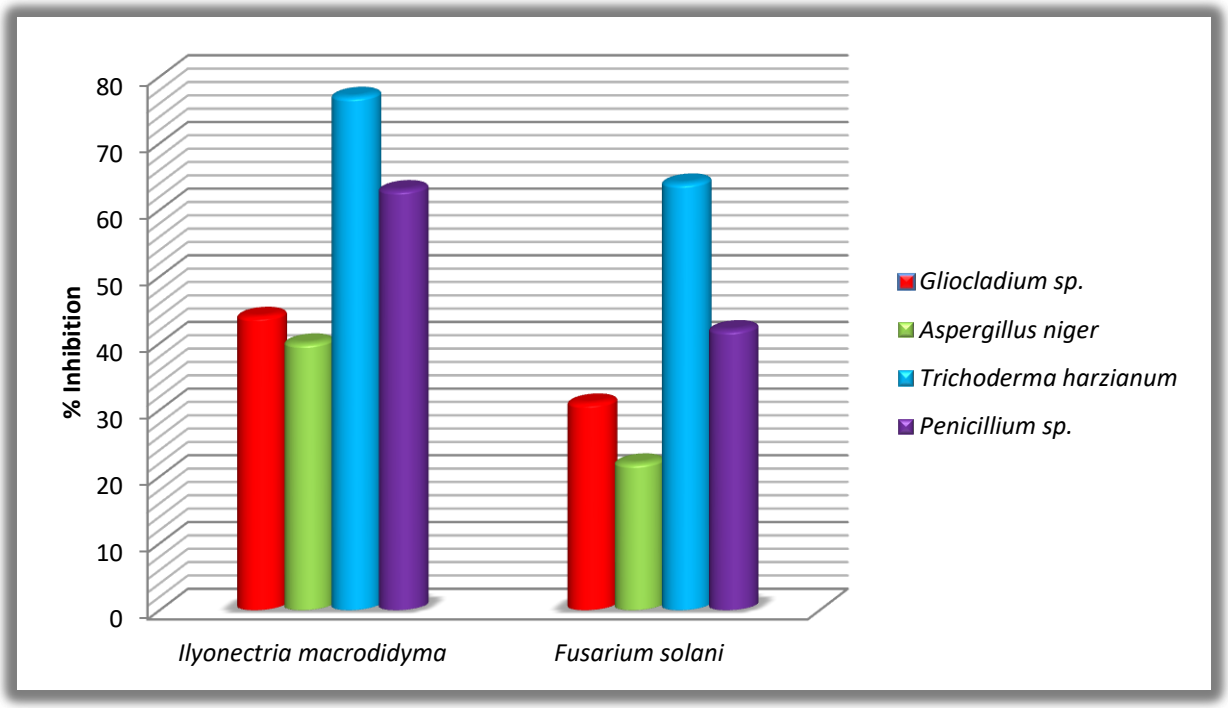

Fig. (5): Inhibition percentage and Bells scale class of Anatagonistics fungi against pathogens using dual culture method

The results of dual culture method revealed that *T. harzianum* completely overgrew the pathogen *I. macrodidyma* and was placed in class1 according to Bells scale (Table 2), whereas *Penicillium sp.* parasitized the tested pathogen up to two-thirds of the medium and were placed in class 2 and remaining antagonists placed in class 3 according to Bells scale. In case of *F. solani*, the antagonists *T. harzianum* overgrew up to two thirds of the medium and

were placed in class 2, while the pathogen colonized at least two-thirds of the medium and overgrew the *Gliocladium sp* colony and was placed in class 4. *Penicillium sp*. and *Aspergillus sp.* colonized one-half of the medium and was placed in class 3. *T. harzianum* has varying levels of growth inhibition toward the *I.macrodidyma* pathogen species tested in vitro compared to the other species. This agrees with (van Jaarsveld et al., 2021).

Table (2): Bells scale class of antagonistic fungi against pathogens using dual culture method

Pathogenic Fungi	Antagonistic fungi			
	Gliocladium sp.	*Aspergillus niger*	*Trichoderma harzianum*	*Penicillium sp.*
Ilyonecteria macrodidyma	3*	3	1	2
Fusarium solani	4	4	2	3

*Bale scale:- 1: the antagonist was completely overgrew the pathogen and covered the medium surface. 2: the antagonist overgrew at least 2/3 of the medium surface. 3, both the antagonist and the pathogen expanded on one half of the medium surface (greater than 1/3 and smaller than 2/3) and none of the organisms appeared to control each other.4: The pathogen colonized at least 2/3 of the medium surface and appeared with the stand overtaking. 5: the growth of the pathogen against the antagonist was successful and the entire surface of the medium was colonized.

In vitro parasitism of *G. catenulatum* against fungal plant pathogens indicated that the antagonist may destroys the mitochondrial cells of *Fusarium spp* by direct interaction without penetration, causing break down of effected cells or disintegrated (Huang, 1978). Win et al., (2021) revealed the involvement of chitinase and b-1, 3-glucanase in the degradation of fungal cell walls because of these two products are the major components of most fungal cell walls. It was also observed that the isolated *Penicillium sp*. showed strong antifungal activity in dual-culture experiment with *F. solani* and *I.macrodidyma*. This antifungal activity of *Penicillium sp.* against *Fusarium* species have has been reported previously by Elkhayat and Goda, (2017). Biological control has some limitations over the seasonal and environmental conditions specific for any given agro-ecosystem. Therefore, microbial metabolites involved in antagonistic activity may exhibit a difference in quantity as well as efficiency as produced under in vitro and in vivo conditions, affecting the apparent reduction in pathogen infectivity (Kohl et al., 2019).

CONCLUSION

Disease symptoms of wilting and dieback were found on olive trees during the field survey in Qasrok, Sharia and Sartank locations which represent the main area of olive production at Duhok province. The predominant isolated fungi in this study were *Fusarium solani* and *Ilyoncteria macrodidyma*. Pathogenicity test showed the high level of olive roots and aerial parts infection by both tested fungi. In vitro efficacy of soil antagonistic fungi observed the most effect isolates able to inhibit the mycelial growth of both pathogenic fungi were *Trichoderma harzianum* and *Penicillium sp.*

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دە ست نیشانکرنا توندیا توشبونا رە هین نە مامکین زیتینا ب کە رویی Fusarium solani و Ilyonectria macrodidyma وکارتیکرنا کە روین ناف ئاخی ل دژی توشبونی

پوختە

لێکۆلینەک هاتە ئەنجامدان لسەر شەش کێلگێت زەیتینا ل سێ جهان (سەرتەنگ و شاریا و قەسروک) ل پاریزگە ها دهوکی. چرترین پلا نەخوشیێ بجرمیسن و مرنا دارین زە یتینا هاتە تومارکن (٤٠٪) و رێژەیا توشبویا (٨٣٪)ل شاریا ،لێ رێژەیا هەرە کێمتر ل سەرتەنگ هاتیە تومارکرن کو گەهشتیە ٣٥٪.کە روین هەرە زیدە هاتینە دهست نیشانکرن د ڤە لێکولینێ دا ژ جورێ Fusarium solani و Ilyonectria macrodidyma بوون. کە روی F. solani بە رزترین کە رو بو هاتیە دە ستنیشانکرن ب رێژەیا(53.3%) ل قەسروکێ ، لێ کە روی I. macrodidyma رێژەیا هەرە کێمتر بوو کو گەهشتیە (16.6٪) . کە روا F. solani ب رێژەیا 29.4٪ ، 27.2% ل شاریا و سەرتەنگ هاتە دیتن. ریزا توشبونی ب F. solani 22.66% و 40.33% و 67% بوو ل هیفین ئایاری و تموز وایلولی . وریژا توشبونی ب I.Macrodidyma 24.67% و 31% و 45.33% ل هە مان دە م و لدویف ئیك. پشتی پێنج مەها شیانا نەخوشیێ رێژەیەکا بەرز ژ توشبونێ دیارکر ب کە روا F. solaniکو رێژەیا١٠٠٪ ئەنجامدا و چربونا توشبونێ ٦٨.٢٠٪ .. نزمبون ئانکو کێمبونا کێشەیا تەر یان هشک ژ بەشێ بلند یان کومەیا کەسک ژ رووەکا ٣٦.٦٤٪ و ٧٥.٢٥٪ و د کومەیا رەهان نزمبون و کێمبون ب رێژەیا ٥٩.٢٩٪ و ٩٩.٥٠ ٪ لدویف ئێک . لێ د کە روا I macrodidyma. رێژەیا توشبونێ ١٠٠٪ و چربونا توشبونێ ٧٠.٤٦٪ د کومبونا رەهان دا . نزمبون ئانکو کێمبونا کێشەیا تەر یان هشک ژ بەشێ بلند یان کومەیا کەسک ژ رووەکا ٢٨.١٣٪ و ١٨.٨٠٪ ، کومەیا رەهان نزمبون و کێمبون ب رێژەیا ٣٤.٦٦٪ و ٢٩.٣٢٪ لدویف ئێکڤە

Trichoderma harzinum پلەز گەشبونا کە روا Fusarium solani راوستاند و کارتێکرنا وی کێمکر بریژەیا ٦٤٪ لێGliocladium spرێژەیا گەشبونا 31٪ کیمکرو کە رو ژ جورێ Aspergillus sp بریژەیا ٤٢٪. کارتێکرنەکا بە رز هاتە نیشانکرنا دژی کە رو I.macrdidyma بریکا کە روا Trichoderam harzianum بریژەیا ٧٧٪ . ئەنجامێت چاندنا دوینی دیاکر کە روTrichoderma harzianum گەشەبونا وێ مەزن بو و بەر بەلاڤ بو و کاریگەریێ ئێشێ ب I. macrodidyma نخافت بتمامی و کرە د رێزبەندیا ١الدویف پیڤەرێن Bells ،لێ کە رو Penicillium sp دژاتیەک دیارکر هەمبەری کارتێکەرێ ل ئەگەرا ئێشێ کر هەتا گەهشتیە نیڤێ و هاتە دانان د رێزبەندیا ٢ . هەروەسا ئەنجامێت چاندنا دوینی دیاکر کە رو بایوئجنت T. harzianumگەشەبونا وێ مەزن بو و بەر بەلاڤ بو و کاریگەریێ ئێشێ ب F. solani نخافت و کرە د رێزبەندیا 2لدویف پیڤەرێن Bells ،لێ کە رو Gliocaldium sp و Aspergillus niger دژاتیە کا کیم هەمبەر کارتێکەرێ ئێشێ کر و گەهشتە کیم تر ژ نیڤێ و هاتە دانان د رێزبەندیا 4. کە روی Penicillium sp هاتە دریزبە ندیا3.

اختبار القدرة الإمراضية للفطرين *Fusarium solani* و *Ilyoncteria macrodidyma* على جذور شتلات الزيتون وبيان الفعالية التضادية للفطريات المعزولة من التربة ضد العدوى

الخلاصة

تم اجراء مسح حقلي على ستة حقول زيتون بشكل عشوائي في ثلاثة مواقع (سرتنك ، شاريا وقسروك) في محافظة دهوك. سجلت أعلى شدة ونسبة ظهور لاعراض الذبول والموت التراجعي على اغصان الزيتون في منطقة شاريا وبشدة ونسبة اصابة بلغت (83% ،40٪ ، 83% على التوالي في حين أقل شدة إصابة كانت في منطقة سرتنك وبنسبة 35٪ .الفطريات السائدة في هذه الدراسة هي *Fusarium solani* و *Ilyonectria macrodidyma*. ظهرت اختلافات للعزلات الفطريه في المواقع المذكورة اعلاه *F. solani* . كان الاكثر تكرارا بنسبة (53.3 %) في قسروك ، بينما *I. macrodidyma* سجل اقل تكرار (16.6%). الفطر *F. solani* سجل بنسبة 29.4٪ و 27.2٪ في شاريا وسرتنك على التوالي. أظهرت شتلات الزيتون التي تم تلقيحها ب الفطر . *I macrodidyma*تلونًا واضحًا ولونًا بنيًا مع ظهوراصفرار بالاوراق وموت حواف الاوراق ، التفاف الاوراق والموت التراجعي للاغضان . وكذلك تتسبب في موت منطقة التاج والاوعيه الجذريه او الاوعية الناقلة ، وتكون تلك المنطقه غائرة مع اختزال في الجذور الثانوية. الشتلات المصابة بالفطر *F. solani* اظهرت اصفرار الاوراق، موت الاغضان ، وتظهر الاوراق باللون البني عند القاعدة ، التاج ، والساق ، وكذلك مع تقدم الاصابه تتغير الى اللون البني وتغير لون الاوعية الناقلة وبذلك تؤدي الى موت الشتلات. شدة الاصابة او المرض في *F. solani* في الجزء العلوي من الشتلات او الاجزاء الهوائية في شتلات الزيتون خلال شهر ايار ،حزيران و ايلول كان 20.66%, 40.33% و 67% على التوالي . بينما كانت شدة الاصابة 24.67%, 31%, 45.33% على التوالي للفطر *I. macrodidyma* .

أظهرت القدرة المرضية بعد خمسة أشهر إصابة عاليةاو شديدة ب الفطر *F. solani* وكانت نسبة الاصابة (100%) وشدة الاصابة (68.20%).الانخفاض او النقص الحاصل في الوزن الرطب والوزن الجاف للجزء العلوي او المجموع الخضري من الشتلات كان 36.64% و 25.75% وفي المجموع الجذري كان الانخفاض او النقص الحاصل بنسبة 59.29% و 50.99% على التوالي *I. macrodidyma* .كانت نسبة الاصابة (100%) وشدة الاصابة (46.70%) في المجموع الجذري . الانخفاض او النقص الحاصل في الوزن الرطب والوزن الجاف للجزء العلوي او المجموع الخضري من الشتلات كان 18.23%, 18.8%، بينما كان الانخفاض او النقص الحاصل في المجموع الجذري 34.66%,29.32% على التوالي . اوقف الفطر المضاد *Trichoderma harzianum* بسرعة نمو الممرض *F. solani* واظهر تثبيطا بمقدار (64%) ، بينما *Gliocladium sp* اظهر تثبيطا للنمو الشعاعي للفطر *F. solani* بنسبة (31%) والفطر *Aspergillus niger* بنسبة (22%) على التوالي . ولوحظ تثبيط عالي للفطر *I. macrodidyma* في المختبر بواسطة الفطر *Trichoderma harzianum* بنسبة (77%) . اظهرت نتائج الاستزراع المزدوج أن الفطر *Trichoderma harzianum* قد نما بشكل مفرط وغطى العامل الممرض . *I macrodidyma* بشكل كامل وتم وضعه بالفئة 1 وفقا لمقياس Bells، بينما الفطر *Penicillium sp* اظهر تطفلا ضد العامل الممرض وصل حتى ثلثي الوسط وتم وضعه بالفئة 2. اظهرالفطر الممرض *F. solani* تطفلا مفرطا على الفطر *Trichoderma harzianum* حتى وصل ثلثي الوسط وتم وضعه بالفئة 2. بينما استعمر العامل الممرض ما لا يقل عن ثلثي الوسط ونمو مستعمرة الفطر *Gliocladium sp* وتم وضعها في الفئة 4. *Penicillium sp* و *Aspergillus niger* استعمرت نصف الوسط وتم وضعها في الفئة 3.