

SUPPRESSION OF THE PATHOGEN OF FUSARIUM WILT *FUSARIUM OXYSPORUM* F.SP. *LYCOPERSICI* ON TOMATO BY INTERCROPPING WITH GARLIC

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ABSTRACT

This study aimed to investigate and evaluate the efficiency of intercropping garlic plants with tomato and their root secretions in controlling the cause of Fusarium wilt disease by using alternative methods to the use of chemical pesticides, and the experiment was carried out in the College of Agricultural Engineering Sciences/University of Baghdad. Garlic root secretions were extracted and their effectiveness in inhibiting radial growth was studied at several concentrations. The concentration of 30 $\mu\text{l.ml}^{-1}$ showed the highest rate of inhibition, reaching 96.3%, compared to the control treatment, which amounted to 0%, and this result confirmed the ability of garlic root secretions to inhibit pathogenic fungi. The effect of planting garlic with tomatoes on plant growth-promoting rhizobacteria (PGPR) populations was also studied, and an increase was found in the numbers of these beneficial bacteria. Intercropping significantly, in contrast to monocropping, which increased by a small percentage in the fruit-setting stage and then continued to decrease in the late fruit stage. In the pot experiment, the results of using two treatments of intercropping garlic with tomato plants and watering with garlic root secretions showed a significant reduction in the percentage of infection and disease index, reaching 58.3% and 25%, respectively. The results showed the effectiveness of the treatments used in increasing the concentrations of defensive compounds such as the enzymes Peroxidase, PAL, and the content of phenols, as the FGR treatment recorded an increase in the induction and accumulation of these compounds when detected in tomato leaves, which amounted to 39 Δ absorbance.min⁻¹.g⁻¹, 31 nM.mi⁻¹.g⁻¹, and 5.47 mg of catechol.g⁻¹, respectively.

Keywords: induce systemic resistance, peroxidase, pal, phenols, chlorophyll, pgpr.

*Part of Ph.D. dissertation of the 1th auther.



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INTRODUCTION

The tomato (*Solanum lycopersicum* L.) plant is a widespread vegetable that belongs to the Solanaceae family, which includes more than 3,000 species, this crop can be widely grown and consumed all over the world due to its high nutritional value and is the second favorite and most consumed vegetable crop after potatoes in the world (Abdallah, et al. 2020; Siddiqui et al. 2020). Global tomato production faces many challenges, especially biotic factors such as fungi, bacteria, nematodes, and viruses, and other abiotic factors such as salinity, drought, waterlogging, cold, and heat stress, which adversely leads to

a reduction in plant growth and productivity (Abdul-Hasan et al. 2016; Ahmad et al. 2013; Al-Tamimi et al. 2020; Singh et al. 2020; Zandalinas et al. 2018). Fusarium wilt is an economically important disease that affects tomato plants wherever they are grown, whether in the field or in greenhouses, and is caused by the pathogen *Fusarium oxysporum* f.sp. *lycopersici* (Fol) (Al-hamiri et al. 2014; Borisade et al. 2018). The disease affects the quality and productivity of fruits at a rate ranging between 50% to 60%, and Losses due to infection with the disease can reach 100% for susceptible varieties when soil conditions and temperatures are suitable (Bawa, 2016;

Liu et al. 2014). The plant is infected at all ages, starting with the seeds rotting and not germinating, then newly developing seedlings are infected. When infected, they die immediately, causing damping off disease. In adult plants, the pathogenic fungus FOL enters the epidermis of the root, subsequently spreads through the vascular tissue, and colonizes the xylem vessels, leading to vessel occlusion and causing severe water stress. Since agriculture is the mainstay of a developing economy to provide food and a better life, the field of agriculture faces a wide range of challenges, including unpredictable climate change, soil pollution with various harmful environmental pollutants such as fertilizers, pesticides, and pathogens and a significant increase in demand for food with the world population are increasing (Pareek Liu et al. 2017). A recent United Nations report predicted that the world population will reach 8.5 billion by 2030 and about 9 billion by 2050, in order to meet the demands of a relentlessly growing population, there is an urgent need to increase food production by more than 50%, to deal with these issues, technologies and strategies are constantly evolving, given this, one such step that has been taken is the introduction of Intercropping to revolutionize modern farming practices. Intercropping is an example of sustainable agricultural systems and a way to increase diversity in the agroecosystem and protect plants (Bedoussac et al. 2018; Boudreau, 2013). The agricultural sector, as an important economic activity in different societies, requires coherent planning in order to achieve development and confront crises, Continuous cultivation of the same crop may cause an imbalance of nutrients in the soil and lead to a rise in pH and secondary salinity. This practice also often leads to the accumulation of toxic substances of harmful microorganisms, which cause damage to crop roots (Li et al. 2020; Xiao et al. 2012). Reasonable intercropping can address many of these problems and increase overall crop growth and production (Chen et al. 2019; Jafar et al. 2021; Joshi, 2018). Intercropping is defined as a multiple cropping system, meaning planting two or more crops in the same field during the growing season, and it is

a means of increasing diversity in the agricultural ecosystem. Increasing environmental balance, increasing the use of resources from the quantity and quality of products, and reducing damage caused by pests, diseases, and weeds (El-Anany et al. 2021; Joshi, 2018). Thus, intercropping is considered an effective, inexpensive, and environmentally friendly alternative. Because tomatoes are one of the most common vegetable plants grown in greenhouses in Iraq, but the obstacles to their cultivation have become a major problem, intercropping of tomato plants with garlic plants is applied, as intercropping is an effective, inexpensive and environmentally friendly alternative in suppressing the cause of Fusarium wilt disease caused by the fungus Fol, and tested its efficiency in inducing induced systemic resistance and increasing growth parameters and crop productivity.

MATERIALS AND METHODS

Fungal pathogen and Inoculation

preparation: The fungal isolate from the pathogen Fol was isolated, phenotypically and genetically identified, and deposited in GenBank under accession number (OQ244516). The fungal inoculum was prepared from the 6-day-old pathogenic fungus Fol according to the method suggested by Taylor et al. (2013) and modified by Aamir et al. (2019). Briefly, the Petri dishes containing the culture were suspended with sterile distilled water. Spores were gently collected using a glass spreader, and the heterogeneous suspension was filtered using a cloth (gauze) to remove the fungal mat. The numbers of spores were calculated using a hemocytometer, and the filtered suspension was diluted with sterile distilled water to reach a concentration of 2×10^6 spores.ml⁻¹.

Effect of garlic root secretions on fungal

growth FOL: Took garlic cloves and sterilized it with a sterile solution (Sodium hypochlorite) at a concentration of 1% (free chlorine) for two minutes and then washed it several times with sterile distilled water to remove the effect of the sterilizing substance. The Garlic Clove was placed on a plastic clip with small holes, which was sterilized with 70% ethanol. Place the sterilized garlic cloves

on it and place a sterile vial bottle (capacity 50 ml) containing sterile distilled water under the clamp, as the water comes into contact with the garlic cloves to ensure their germination, then, 25 days after germination, the water containing the secretions was taken into the centrifuge (at a speed of 3000 rpm) to precipitate the impurities in it. It was filtered using Whatman No. 1 filter papers, then the filtrate was passed through the fine filter (Millipore 0.22 μ l). Root exudates at -20°C until use (Al-shafiee and Dewan, 2015). Then tested several concentrations to inhibit the pathogenic fungus (10, 15, 20, 25 and 30 $\mu\text{l.ml}^{-1}$) by taking 1, 1.5, 2, 2.5 and 3 ml of water containing the filters and adding it to 100 ml of PDA nutrient medium before pouring it into the dishes. In addition to the control treatment, which added sterile distilled water. The center of each dish was inoculated with a 0.5 cm diameter disk of the pathogenic fungus Fol at 6 days old with three replicates, then the dishes were incubated at a temperature of $25\pm 2^{\circ}\text{C}$. The percentage of fungal inactivation after 6 days was calculated on the basis of the percentage of inhibition of radial growth (IRG), according to as in the following equation (Hermida-Montero et al. 2019):

$$\text{IRG} = \left(\frac{R_1 - R_2}{R_1} \right) \times 100$$

As R_1 : is the radial growth in the control treatment

R_2 : is the radial growth in a treatment with the presence of the pathogenic fungus Fol

The effect of growing garlic with tomatoes on bacterial populations

Soil samples were collected at different growth stages of the tomato crop, at a distance of 5 cm from the tomato plant and a depth of 15 cm: before tomato transplanting (BTT), fruit set stage (FSS), early fruit stage (EFS), middle fruit stage (MFS) and late fruit stage (LFS), respectively. The collected samples were mixed well to create a composite sample of each treatment for each replicate. Samples were stored at 4°C for Agriculturally Important Microbes (AIMs) analysis. 5 g of each composite sample was weighed and placed in a 200 ml glass beaker with 45 ml of sterile distilled water. The bottle was shaken

for 30 minutes on an electric shaker, then the homogeneous solution was serially diluted to 10^{-5} . Solutions were used at a dilution of 10^{-5} to inoculate dishes containing Nutrient Agar medium (NA), and bacterial populations were counted after two days of incubation at 28°C with three replicates for each treatment. The number of bacterial colonies in the medium was measured using the standard dilution plate method in terms of colony forming units (CFU).

Experiment design: The mixed soil was sterilized with a steam sterilizer (Autoclave) for two consecutive times, after which it was distributed in plastic pots with a diameter of 19 cm. I planted 30-day-old tomato seedlings (Newton var.) in the pot, after dipping them in the spore suspension of the pathogenic fungus (at a concentration of 2×10^6 spores. ml^{-1}), for half an hour, and after three days, seedlings of local garlic were planted, with approximately 4 garlic cloves per pot, and 50 ml of garlic root secretions were added with the rest of the treatments. The experiment was carried out with three replicates for each treatment as follows:

Control, Pathogen Fol (F), Garlic plant (G), Garlic root secretions (R), Pathogen + garlic plant (FG), Pathogen + garlic root secretions (FR), Pathogen+ garlic plant + root secretions (FGR).

Disease index and percentage of infection

The disease index and percentage of infection were calculated 35 days after adding the fungal inoculum in the pot experiment. The infection rate and disease index were calculated after 90 days in the greenhouse experiment. The infection percentage was calculated according to the following equation:

$$\text{Percentage of infection} = \frac{\text{No. infected plants}}{\text{Total No. tested plants total}} \times 100$$

The scale was used according to (Fujikawa et al. 2021) with some modifications:

0: The plant is healthy, 1: More than 0-25% of leaves are yellow or wilted, 2: More than 26-50% of the leaves are yellow or wilted, 3: More than 51-75% of the leaves are yellow or wilted, 4: More than 76-100% of the leaves are yellow or wilted, 5: Plant drying out and dying. and the disease index (DI) reported by

(Chen et al. 2019) according to the following equation:

$$DI = \left[\frac{\sum(\text{rating score} \times \text{number of plants rated})}{(\text{total number of plants} \times 5)} \right] \times 100$$

Biochemical evaluation of plant defense response in stimulating induced systemic resistance:

Biochemical evaluation was performed by measuring the amounts of peroxidase (Hammerschmidt et al. 1082), PAL enzyme (Dickerson et al. 1984), and total phenol content (Pouratashi and Irvani, 2012) produced and accumulated in leaf tissues collected from all treated and control samples collected at intervals Different times after 7, 14 and 21 days from adding the pathogen.

Plant chlorophyll content and some growth parameters:

The chlorophyll content of tomato plant leaves was calculated 30 days after adding the pathogen using the SPAD 205 Plus device to estimate the amount of chlorophyll in the plant leaves in a non-destructive way from the middle of the plant, and the length of the shoot and root system 40 days after adding the fungal inoculum. At the end of the experiment, the wet and dry weight was calculated.

Statistical analysis: The data were subjected to statistical analysis using the SAS statistical program for analysis of variance (ANOVA),

and a complete randomized design (CRD) was used in implementing the experiment, and the means were compared according to the least significant difference (L.S.D.) at the probability level 0.05 (SAS, 2012).

RESULTS AND DISCUSSIONS

The effect of garlic root secretions on the growth of the pathogenic fungus *Fusarium oxysporum* f.sp. *lycopersici*:

Five concentrations of garlic root secretions (10, 15, 20, 25, and 30 $\mu\text{l.ml}^{-1}$) were evaluated and the results showed the effect of these secretions on the radial growth of the pathogenic fungus Fol on the PDA nutrient medium, as growth decreased depending on the concentration used. However, the exact mechanism of how these secretions reduce pathogen growth is not clear to us. A clear inhibition and sharp decrease in the radial growth of the pathogenic fungus was observed at the concentration of 30 $\mu\text{l.ml}^{-1}$, as the inhibition rate reached 96.3%, while the concentrations of 25, 20, and 15 $\mu\text{l.ml}^{-1}$ showed an inhibition rate of 72.2, 52.5 and 34%, respectively (Fig. 1), while the concentration of 10 $\mu\text{l.ml}^{-1}$ showed a lower rate of inhibition than the previous concentrations, reaching 17% 6 days after the completion of growth of the fungus Fol in the comparison treatment.

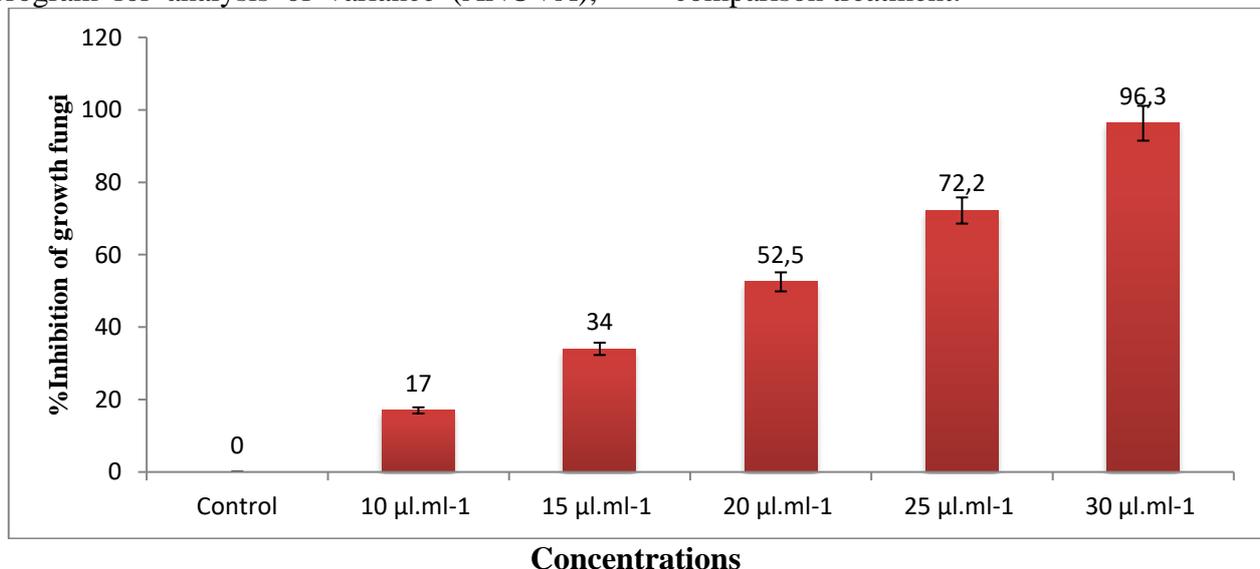


Fig. 1. Percentage inhibition of radial growth of pathogenic fungi by garlic root secretions, L.S.D 0.05= 3.9

According to this study, the antifungal compounds secreted from garlic roots played a role in directly inhibiting the pathogenic

fungus on the PDA nutrient media. Mentioned (Yang and Wu, 2011), that plant root secretions contain phenolic compounds that are excreted into the soil. Indicated (Zhang et

al. 2013) that garlic is able to produce antioxidant compounds and volatile substances secreted from the roots and have an effective role in stopping the growth and reproduction of pathogenic fungi, including dimethyl disulfide, 2-methyl-2-pentenal, dipropyl disulfide and dipropyl trisulfide; In another study conducted by Li et al. (2019), it was shown that the rhizomes of *Atractylodes lancea* have the ability to produce 21 volatile substances, the most important of which are terpenes and aromatic hydrocarbons.

The effect of growing garlic with tomatoes on bacterial populations

The number of bacteria PGPR appeared to change during different growth stages of tomato, but the trend differed between the monoculture and intercropping treatments (Fig. 2). In the monoculture, the number of

bacteria increased from 4.3×10^5 cfu.g⁻¹ before tomato planting to 9×10^5 cfu.g⁻¹ at the fruit-setting stage and to 8×10^5 cfu.g⁻¹ at the early fruit stage, then decreased sharply to 4.6×10^5 cfu.g⁻¹ in the middle fruit stage and 4.6×10^5 cfu.g⁻¹ in the late fruit stage. In the intercropping treatment, the number of bacteria gradually increased from 4×10^8 cfu.g⁻¹ to 13×10^8 cfu.g⁻¹ at the fruit-setting stage during strong growth of garlic, then decreased proportionally to 16×10^8 cfu.g⁻¹ at the early fruit stage. When the garlic was harvested. Then an increase in the number of bacteria was observed to 18.6×10^8 cfu.g⁻¹ in the late fruit stage. Therefore, intercropping garlic increased the number of bacteria in the soil, especially during the growth stages of tomato fruits.

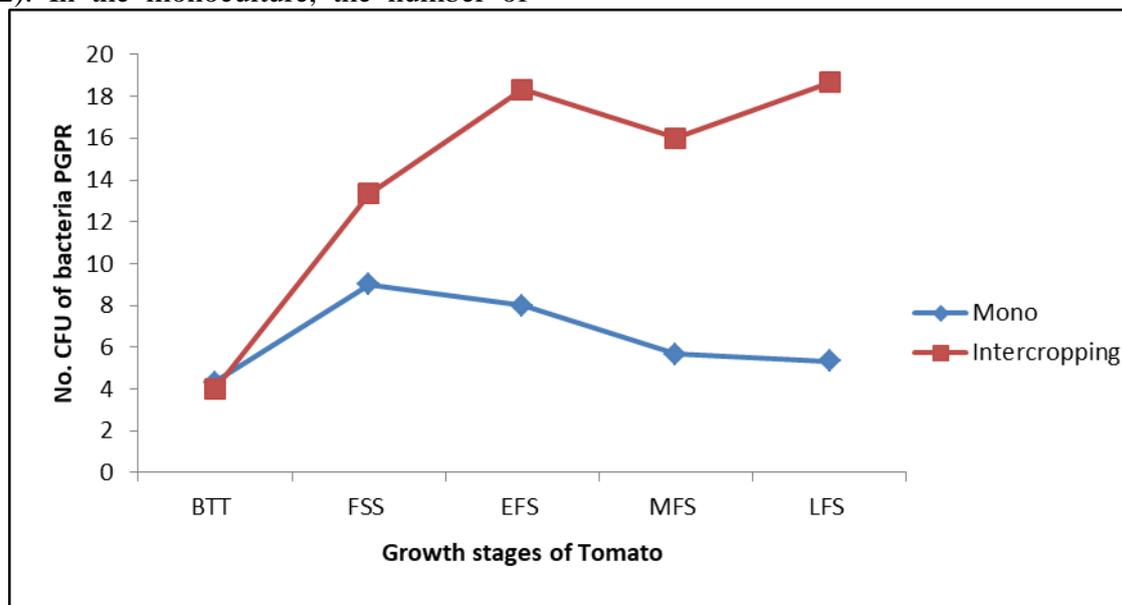


Figure 2. The effect of growing garlic with tomatoes on bacterial populations

Root exudates affect the diversity and number of agriculturally important organisms (AIMs), such as PGPR that live in the soil in the root zone of intercropped plants. This difference is attributed to the quantity and type of these exudates, because of the roles these organisms play in providing protection for the plant from pathogens and enhancing its safety by inducing systemic resistance, and their important effects on vegetative and root growth and increasing productivity. And Li et al. (2018) mentioned in their study the effect of intercropping garlic with tomatoes on the total number of beneficial microorganisms,

including bacteria, fungi, and actinomycetes. Also, (Li et al. 2019) showed in their study the below-ground effects of cultivating peanuts with the Chinese medicinal herb *Atractylodes lancea* on both the fungal and bacterial communities in the peanut root zone, as closed cultivation systems were established to study the role of volatile substances and secretions emerging from the rhizomes. *A.lancea* in fungal and bacterial communities. The interaction led to a significant change in the composition of the fungal and bacterial community in the peanut rhizosphere, and also demonstrated the effect of volatiles from

A.lancea rhizomes on the composition of the soil fungal and bacterial community.

Disease index and percentage of infection of fusarium wilt: Analysis of the data in this experiment showed that all treatments significantly reduced the disease index and percentage of infection, as disease symptoms were observed on the leaves with slight yellowing starting from the bottom of the plant to typical symptoms such as yellowing of most leaves, stunting, or complete wilting, and the matter may lead to death and drying of the plant in the inoculated treatments. With pathogenic fungi only compared to unvaccinated plants that appear with healthy growth and green leaves. This is due to the ability of the pathogen to infect due to its powerful mechanisms against the plant host. Intercropping garlic in the presence of the

pathogenic fungus (FG), and treatment with garlic root secretions in the presence of the pathogenic fungus (FR) showed effectiveness in protecting the plant from Fusarium wilt disease and reducing the infection rate and disease index by 91.6% and 51.6%. 91.6% and 55%, respectively (Fig. 3). While the combination treatment between planting garlic plants and root exudates in the presence of the pathogen (FGR) led to a better reduction in the infection rate and disease index and a significant reduction in preventing the occurrence of Fusarium wilt disease, as the infection percentage and disease index reached 58.3% and 25%, respectively. Compared to the treatment of the pathogenic fungus (F), which showed an infection rate and disease index of tomato plants of 100% and 88.3%.

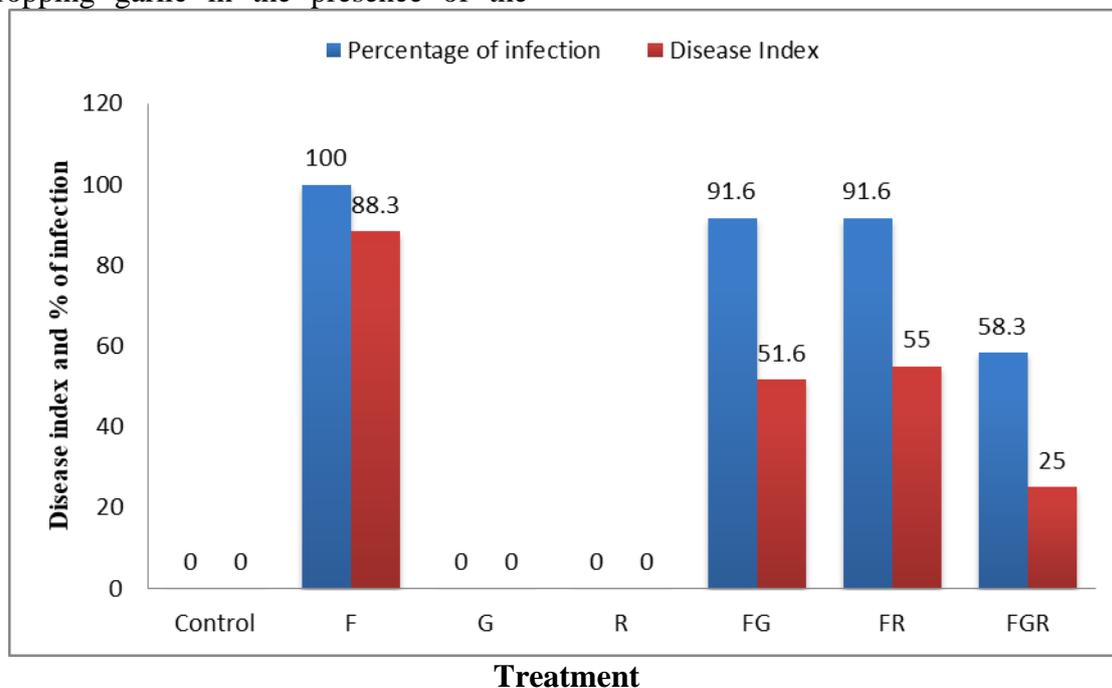


Fig. 3: Percentage of infection and disease index of Fusarium wilt on tomatoes in pots L.S.D 0.05= Index Disease 13.1, % of Infection 16. 5

Control, Pathogen Fol (F), Garlic plant (G), Garlic root secretions (R), Pathogen + garlic plant (FG), Pathogen + garlic root secretions (FR), Pathogen+ garlic plant + root secretions (FGR).

Characteristic wilt symptoms occur due to blockage of transport vessels resulting from the assembly of fungal hyphae and a combination of host-pathogen interaction such as the release of toxins, gums, gels, and formation of thyloses (López-Zapata et al. 2021). The pathogenic fungus produces

secondary metabolites such as Fusaric acid, Fumonisin, Lycopersin, and Dehydrofusaric acid, and the effects of the toxin are associated with decreased photosynthesis, lipid peroxidation, wilting, leaf necrosis, and cell death (Singh et al. 2017), as well as its the ability to produce many enzymes that break down the cell wall, allowing penetration and colonization of root tissues (McGovern, 2015). As the disease progresses, apical growth stops, and the fruits

are few or do not develop. If the main stem is cut lengthwise, dark brown discoloration can be seen. Brown discoloration in the vascular system is a feature of the disease and can generally be used to identify the disease. The results also showed a decrease in the severity of Fusarium wilt infection on tomatoes treated with garlic root secretions alone, and this may be attributed to inhibiting the growth of the fungus and inducing resistance. Likewise, seedlings grown with garlic are also attributed to their direct inhibition of the pathogen, as (Ahmad et al. 2013) indicated that garlic is famous for its antioxidant components. For microbes, mainly allicin. Exudates secreted by the root system of garlic can cause obvious effects on soil structure and the environment. This researcher (Zhang et al. 2013) demonstrated the ability of garlic to secrete compounds that inhibit pathogenic fungi and volatile substances such as dimethyl trisulfide, dimethyl disulfide, 2-methyl-2-pentenal, dipropyl disulfide, and dipropyl trisulfide. He explained the effectiveness of these volatile substances and aqueous leachates from leaves and roots. Garlic in suppressing Fusarium wilt disease on bananas caused by the pathogenic fungus *Fusarium oxysporum* f.sp. *cubense* (FOC) when using Chinese chives as an intercropping. In a closed system that mimics the natural environment, volatiles released by the leaves and roots of Chinese chives inhibit the growth of hyphae and the germination of FOC fungal spores. Found Li et al. (2019) that the peanut root rot disease caused by *F.oxysporum* was reduced by intercropping with the medicinal herb *Atractylodes lancea*. Volatile substances emerging from *A.lancea* rhizomes significantly changed the community composition of beneficial fungal and bacterial microorganisms in the peanut root zone, simultaneously significantly inhibiting the growth of pathogenic fungi and improving peanut growth. GC-MS analysis revealed the presence of 21 volatile substances originating from *A.lancea* rhizomes, with terpenes and aromatic hydrocarbons being the most common types.

Induction systemic resistance in tomato plants: The treatments used led to the

stimulation of physiological responses by increasing the production and accumulation of defensive biochemical indicators such as the enzymes Peroxidase (POX) and Phenylalanine ammonia-lyase (PAL) and the phenol content in the tomato plant. Thus, the plant's defenses were strengthened and the entire plant was made more resistant to the pathogen. These were measured after 7 days, days and reached the highest level after 14 days, after which it began a gradual decline, but it remained at a higher level than on the seventh day. Thus, these defensive compounds showed their effective role in stopping and reducing the infection with the pathogen Fol, and this result supported what was mentioned above in that these treatments led to a reduction in the infection rate and the disease index. In the pot experiment (Fig. 4, 5, 6), the fungus pathogenic to unit (F) showed, after 7 days, a slight increase in the concentration of defensive compounds, but after 14 and 21 days it continued to decrease, as POX activity recorded from 18 to 16, then 7 Δ absorbance.min⁻¹.g⁻¹, and PAL activity recorded 5 to 6, then 4 nM.mi⁻¹.g⁻¹, and recorded Phenols activity 0.50 to 0.53 then 0.20 mg of catechol.g⁻¹. Because the disease fungus possesses mechanisms through which it works to stop and disable the plant's defensive response and works to destroy it, thus its concentration decreases as the infection progresses. However, the treatments had an effective role in demonstrating their efficiency in increasing biochemical indicators, as the highest activity was recorded at 14 days of adding the treatments. In individual treatments, such as the intercropping treatment of garlic in the presence of the pathogenic fungus (FG), on day 14, the highest level of induction of POX, PAL, and phenols was recorded. They reached 31, 20 and 3.87, followed by treatments with garlic root secretions in the presence of the pathogenic fungus (FR) with a percentage of 25, 16 and 3. While the FGR combination treatment achieved the highest level recorded on day 14 for the concentration of defensive compounds, reaching 39, 31 And 5.47, respectively.

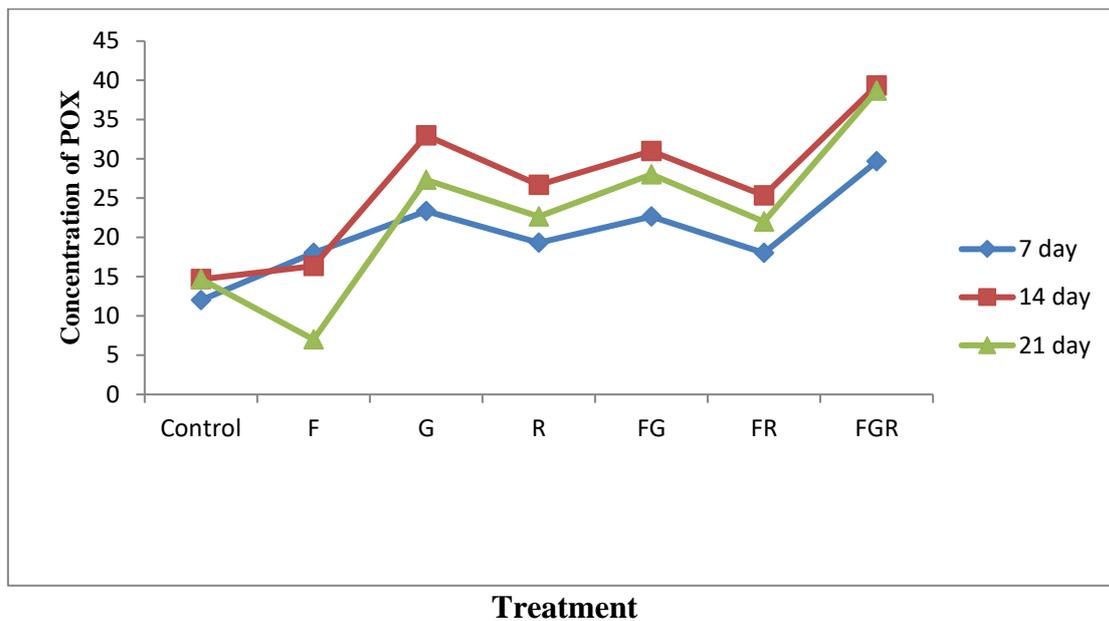


Fig. 4. POX enzyme concentration in pots, L.S.D 0.05 = 2.8, 3.1 and 2.7, respectively.

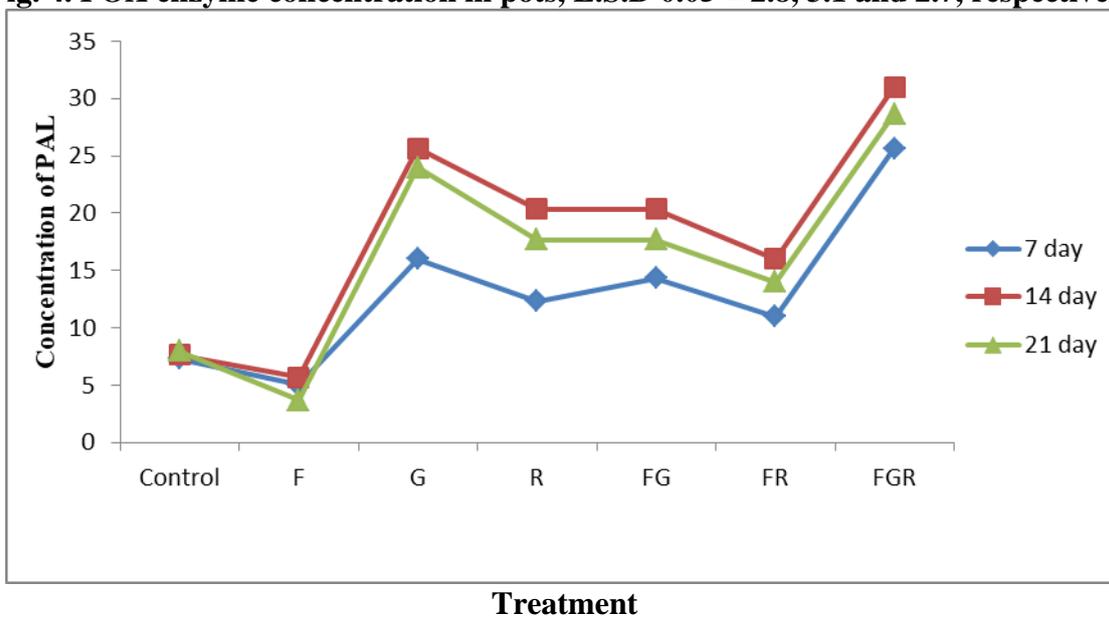


Fig. 5. PAL enzyme concentration in pots, L.S.D 0.05= 2.3, 2.4 and 2.1, respectively

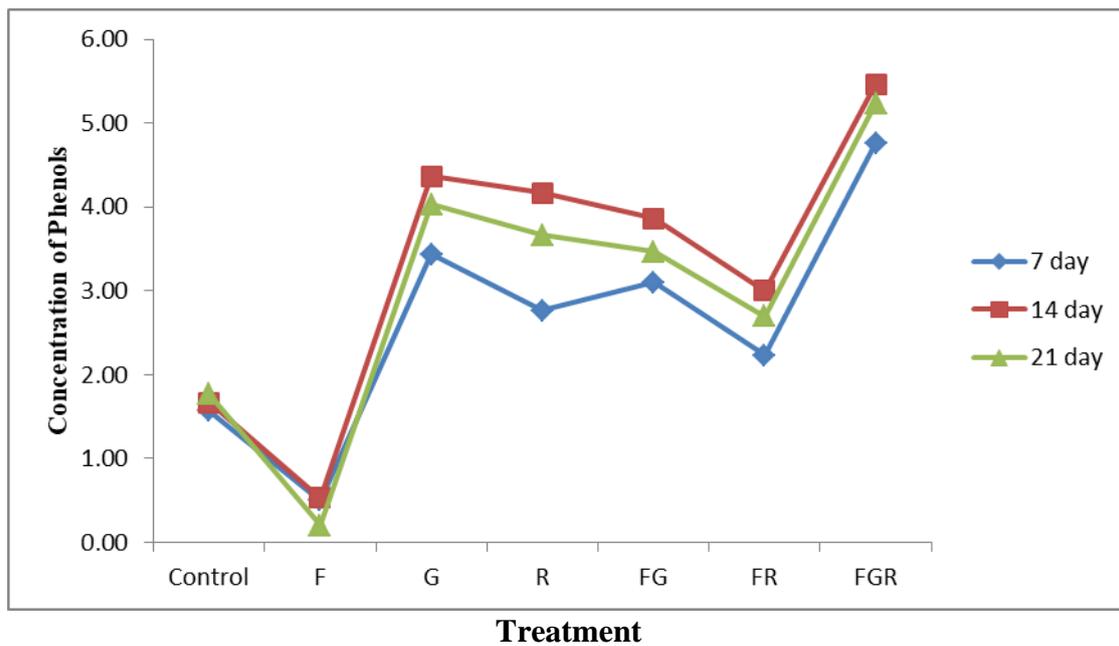


Fig. 6. Concentration of Phenols in pots, L.S.D 0.05= 0.4, 0.5 and 0.9, respectively

Induced systemic resistance (ISR) is a physiological state of stimulated defensive ability in plants caused by biological or chemical stimuli, which protects plant tissues not exposed to the initial attack against future attack by pathogens. The application of environmentally friendly treatments enhances the host's immune defenses against a wide range of pathogens. Plants can recognize microbial- or pathogen-associated molecular patterns (MAMPs or PAMPs), such as fungal chitin. However, pathogens can overcome the first class by suppressing PTI signals or evading recognition. PRRs are expressed by secreting virulence effectors (Rishi et al. 2008; Yu et al. 2022). For this reason, environmentally and health-safe methods have been resorted to treating the plant, such as intercropping of garlic and root secretions, which lead to the creation of a physiological state for the plant host, called priming to stimulate the production and accumulation of defensive compounds such as the enzyme POX and PAL and phenols content. It has been shown that intercropping wheat with watermelon can enhance resistance to *Fusarium wilt* disease in watermelon plants caused by the pathogenic fungus *Fusarium oxysporum* f.sp. *iveum*, as it increases lignin biosynthesis and defense-related gene expression and increases β -1,3-glucanase

activity, in addition, soil microorganisms, whose presence increases with intercropping, can contribute to increasing the resistance of watermelon plants to diseases. Tissue culture experiments also showed that both external addition of wheat root exudates led to increased expression of defense genes such as lignin and total phenolics, and increased β -1,3-glucanase activity in watermelon roots (Li et al. 2020). Showed El-Anany et al. (2021) the effect of extract spray treatments and intercropping of garlic or onion with potato plants on phenolic content and enzymatic activities, all treatments led to a significant increase in total, free and bound phenols. These treatments also stimulated the activities of defense enzymes in the plant such as Chitinase, Polyphenol oxidase, Peroxidase and Phenylalanine ammonia-lyase, thus reducing the incidence of late blight disease caused by the pathogenic fungus *Phytophthora infestans*. The effective role of allicin in reducing the infection of the pathogenic and inhibiting the germination of its spores. Conducted Alizadeh et al. (2023) a study to evaluate the allelopathic effects of wheat as a staple crop and redroot pigweed (*Amaranthus retroflexus* L.), and demonstrated the effectiveness of this evidence of cultivation in inducing systemic resistance and increasing the activity of enzymes such as peroxidase, superoxide dismutase, and catalase in wheat

plants, and the compounds secreted from the roots of the herb were identified using GC-MS. The compounds were terpenoids, phenolic compounds, fatty alcohol, steroids, fatty acids, and alkanes.

Growth indicators of tomato plants

The results showed a significant difference in the growth of tomato plants between the treatments used when intercropping garlic plants (4 cloves) with tomato plants and adding 50 ml of garlic root secretions in 6 weeks of the crop's life. The reason is attributed to the type of treatments and the extent of their effect. All treatments improved the growth. The plant, its health, and vital growth indicators were higher compared to the control treatment. Treatment of garlic plants intercropping with tomato plants in the presence of the pathogenic fungus (FG)

showed a higher index of growth parameters such as root and shoot length, wet and dry weight, and chlorophyll content, reaching 56 cm, 9.5 cm, 55 g, 11.5 g, and 55 SPAD, respectively, compared to the irrigation treatment. Garlic root secretions amounted to 53.2 cm, 8.5 cm, 51.8 g, 10.3 g and 54 SPAD. The best result that could be obtained in the pot experiment was recorded in the combination treatment between planting garlic plants and watering with garlic root secretions in the presence of the pathogenic fungus (FGR), as the shoot length, root shoot length, wet weight, dry weight and the plant's chlorophyll content reached 81.5 cm, 15.1 cm, 72.5 g, 22 g and 91 SPAD respectively, compared to the fungus treatment for unit (F) 21.2 cm, 4.5 cm, 16.5 g, 4.3 g and 27 SPAD (Table 1).

Table 1. Growth indicators of tomato plants in pots

Treatments	Shoot length (cm)	Root length (cm)	Wet weight (g)	Dry weight (g)	Chlorophyll content
Control	60.2	7.4	53.8	10.5	51
F	21.2	4.5	16.5	4.3	27
G	76	12.3	65.5	15.3	88
R	73.5	10.8	61.3	12.5	82
FG	56	9.5	55	11.5	55
FR	53.2	8.5	51.8	10.3	54
FGR	81.5	15.1	72.5	22	91
L.S.D. 0.05	4.49	1.75	3.06	2.15	3.55

Phenotypic characteristics such as plant height, stem thickness, root system length, leaf density, number and weight of fruits, and leaf color; The internal plant's content of elements and protein is evidence of the good growth of the plant and the efficiency of the treatments used in increasing vital growth indicators and protecting the plant from infection with the pathogen Fol. Thus, the treatments used, in addition to their effect on the pathogenic fungi and stimulating plant defenses, have been found to enhance plant nutrition and growth. Among the vital indicators that were measured is chlorophyll, as determining the chlorophyll content in leaves is one of the basic techniques in studying the process of photosynthesis, and chlorophyll is a measure of plant health in agricultural aspects, and since the process of photosynthesis is the basic process during which light energy is absorbed and converted into matter. Organically, the importance of the plant pigment chlorophyll as an intermediary

in converting absorbed solar energy and its activity in the process of photosynthesis and the synthesis of organic substances in plants is extremely important (Murugan et al. 2020; Pareek et al. 2017). Indicated Li et al. (2019) in their study that growing peanuts with the Chinese medicinal herb, *Atractylodes lancea*, led to a reduction in the wilt disease affecting peanuts caused by the soil-borne fungus *F.oxysporum*, and also led to an increase in growth and productivity of peanuts. It has been shown that there is an important role for this herb in increasing the fungal and bacterial communities in the area around the roots, which limits the pathogen and encourages plant growth. The herb's secretions of volatile substances and other secretions serve the same purpose.

CONCLUSION

In conclusion, this study describes the extraction of garlic root exudates and tests their antagonistic efficacy in inhibiting the

pathogen *Fusarium oxysporum* f.sp. *lycopersici*, which causes Fusarium wilt on tomatoes in the laboratory, and studying the role of intercropping and its effect on the beneficial microbial community such as PGPR, which increases its presence in the rhizosphere, as well as studying the efficiency of intercropping garlic and garlic root secretions in controlling the pathogen and inducing defensive compounds such as POX, PAL, and Phenols in induced systemic resistance, as well as the role of these treatments in increasing shoot and root length, wet and dry weight, and chlorophyll content in tomato plant leaves.

CONFLICT OF INTEREST

The authors declare that they have no conflicts of interest.

DECLARATION OF FUND

The authors declare that they have not received a fund.

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قمع مسبب مرض الذبول الفيوزارمي *Fusarium oxysporum f.sp. lycopersici* على الطماطة عن طريق الزراعة

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المستخلص

هدفت الدراسة إلى التحقيق في تقويم كفاءة الزراعة البينية لنباتات الثوم مع الطماطة وافرازات جذورها في السيطرة على مسبب مرض الذبول الفيوزارمي باتباع طرق بديلة عن استعمال المبيدات الكيميائية، وتم تنفيذ التجربة في كلية علوم الهندسة الزراعية / جامعة بغداد. إذ تم استخلاص افرازات جذور نبات الثوم ودراسة فعاليتها في تثبيط النمو الشعاعي بعدة تراكيز، إذ اظهر التركيز 30 مايكرو لتر.مل⁻¹ اعلى نسبة تثبيط بلغت 96.3% مقارنة بمعاملة السيطرة التي بلغت 0%، وهذه النتيجة اكدت قدرة الثوم على تثبيط الفطر الممرض. كذلك تم دراسة تأثير زراعة الثوم مع الطماطة على التجمعات البكتيرية المحفزة لنمو النبات PGPR ووجد تزايد في اعداد هذه البكتريا النافعة في الزراعة البينية بشكل كبير على العكس من الزراعة المنفردة التي زادت بنسبة ضئيلة في مرحلة عقد الثمار وبعدها استمرت بالانخفاض في مرحلة الثمار المتأخرة. في تجربة الاخص، اظهرت نتائج استعمال معالمتي زراعة الثوم بينياً مع نبات الطماطة والسقي بإفرازات جذور الثوم الى خفض نسبة الاصابة ومؤشر المرض بشكل كبير إذ بلغت 58.3% و 25% على التتابع. اوضحت النتائج فعالية المعاملات المستعملة في زيادة تراكيز المركبات الدفاعية مثل انزيمي Peroxidase، PAL، ومحتوى الفينولات إذ سجلت معاملة FGR زيادة في استحثاث وتراكم هذه المركبات في عند الكشف عنها في اوراق نبات الطماطة إذ بلغت 39 $\Delta\text{absorbance} \cdot \text{min}^{-1} \cdot \text{g}^{-1}$ ، 31 $\text{mg of catechol} \cdot \text{g}^{-1}$ و 5.47 $\text{nM} \cdot \text{mi}^{-1} \cdot \text{g}^{-1}$ على التتابع.

الكلمات المفتاحية: المقاومة الجهازية المستحثة، pal، peroxidase، الفينولات، الكلوروفيل، pgpr.

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