Integrated Control of White Rot in Eggplant by using *Pseudomonas flourescens*, Penicillium Commune and *Trichoderma asperellum* in Iraq

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Integrated Control of White Rot in Eggplant by using *Pseudomonas flourescens*, Penicillium Commune and *Trichoderma asperellum* in Iraq

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Abstract. The results of isolation and diagnosis of the fungus growing from Sclerotia and isolated from the infected explants showed that the pathogenic fungus *Sclerotinia sclerotiorum* was isolated. The results of the antagonistic ability test of the biological resistance elements against *S.sclerotiorum* gave a high antagonistic ability compared to the control treatment. The treatment of *P.fluorescens* gave the highest percentage of inhibition against the pathogenic fungus, followed by the treatment of *P.commune* and *T.asperellum*. The results showed the efficiency of biological resistance elements, including *P.fluorescens*, *T.asperellum* and *P.commune*, either singly or in combination with each other, in reducing the percentage of infection severity with *S.sclerotiorum* under plastic house conditions and in the greenhouse, where it amounted between 5.55-50 compared to the control treatment (pathogenic fungi alone), which amounted to 94.44% and under field conditions in the greenhouse, which amounted to 5.55-66.66% compared to the control treatment, which amounted to 83.33%. It provided good protection for eggplant, which was positively reflected on all plant growth parameters, in addition to an increase in the cumulative weight of eggplant fruits.

Keywords. White rot, Sclerotia, *Pseudomonas flourescens.*

1. Introduction

The eggplant (*Solanum melongena* L.) is considered one of the plants of the Solanaceae family. It is one of the important vegetable crops that is grown in open and covered houses to benefit from its fruits that are used for various purposes, including cooking, canning, and pickling, in addition to its other medicinal benefits for patients with asthma, diabetes, and urinary system pain. It also works to reduce cholesterol in the blood [1]. Eggplant is infected with a number of pathogens, especially fungi endemic in the soil. The most important fungus, *Sclerotinia sclerotiorum* (Lib.) DeBary, causes white mold disease, which causes significant economic losses in its production [2]. increasing concern about the impact of pesticides in general and fungicides and herbicides in particular on the environment and human health necessitated the development of alternative agricultural techniques [3]. Due to bio control is one of the most important alternatives available, research focused on it [4]. The fungus is considered one of the most important factors used in biocontrol and the fungus *Penicillium* spp. is one of these biological agents, where some of its types are known for their antagonistic activity against

pathogens by secreting antibiotics and inducing systemic resistance in plants by activating multiple plant defense signals [5]. There are numerous studies confirming that this fungus interacts with the roots of crop plants to promote root growth [6]. Several studies have also shown the efficacy of *Trichoderma* spp. In the control of the fungus that causes white rot *S.sclerotiorum* on different crops (7, 8). In addition to the presence of Plant Growth Promoting Rhizobacteria (PGPR) that colonize the roots of the plant and directly improve its growth and yield. Among these bacteria is the genus *Pseudomonas*, *Bacillus*, and others, where these bacteria contribute to the growth of roots and the ability to antagonize pathogens and the formation of Siderophores and some enzymes such as Chitinase and other compounds such as antibiotics and cyanide. Bacteria are able to build the enzyme ACC deaminase, which lowers the concentration of ethylene, thus stimulating growth. The bacteria also build the enzyme B-1,3-glucanase, improving nutrient absorption and accelerating the onset of stress resistance [9]. Due to the importance of white rot disease on the eggplant plant and its widespread, the research aimed to use some bio control elements to control it and reduce its damage.

2. Materials and Methods

2.1. Isolation and Identification of the Pathogenic fungus S.sclerotiorum

Isolation of pathogenic fungi from the stems of infected plants by collecting the sclerotia formed inside the stems of infected plants. The sclerotia were superficially sterilized with 2% sodium hypochlorite solution prepared by O, Neill [10]. The sclerotia were placed in the sterile dilute solution for 3 minutes. The sclerotia were washed with sterile distilled water for the purpose of removing residuals of the solution and transferred onto a sterile filter paper for the purpose of removing excess water. Then, it was transferred by sterile forceps to plastic Petri dishes with a diameter of 9 cm containing potato dextrose agar (PDA) medium supplemented with the antibiotic Tetracycline at a concentration of 250 mg/liter, after sterilization with an autoclave device (121 ° C at a pressure of 5.1 kg / cm²) for 15 minutes With one stone body per plate The dishes were incubated in the incubator at 24 °C for five days or until the formation of the sclerotia, after which the fungus was identified using the taxonomic keys of the fungus, and the sclerotia were kept until they were used in the subsequent experiments of the study.

2.2. Test the Antagonistic Ability of P. fluorescens Bacteria and T. asperellum, P.commune and humic Acid in Inhibiting the Isolation of the Pathogenic fungus S.sclerotiorum in Labotary and on PDA Medium

The experiment was conducted using the double culture technique to study the antagonistic relationship between the pathogenic fungus *S.sclerotiorum*. To *P.commune* Where a Petri dish containing PDA medium was divided into two equal parts of imaginary diameter, where the center of the first half of the dish was inoculated with a disc diameter of 0.5 cm from the fungal growth of a 5-day-old pathogenic colony, and the center of the other half was inoculated with a disc similar to *T.asperellum*, The same experiment was repeated using the *P .commune* bio-resistant fungi against the pathogenic fungus *S.sclerotiorum*. As for the test of *P.fluorescens* bacteria, the bacteria were prepared on Nutrient broth medium in sterilized 500ml glass flasks in an oxidizer at a temperature of 121°C and a pressure of 1.5 kg/cm^2 for 15 minutes. After that, the medium was inoculated from the bacteria to be prepared by taking a swab using a sterile lube of bacterial growth growing on the previously prepared Nutrient agar medium, at 48 hours of age. The components of the flasks were mixed well and incubated at a temperature of 32 ± 3 °C for 3-4 days, then a series of dilutions (10^{-1} ... 10^{-8}) of the bacterial inoculum was prepared, then the medium of PDA was prepared without adding the antibacterial Tetracycline, The dishes prepared for the experiment were inoculated by taking 1 ml of each dilution of the bacterial inoculum *P.fluorescens* in the form of 4 spots on equal dimensions from the circumference of a circle with a diameter of 4 cm, while moving the dish with a capillary movement to distribute the bacterial inoculum homogeneously for each dish. The dishes were inoculated with a 0.5 cm diameter disc of the pathogenic fungus *S.sclerotiorum* taken from the edge of the colony at 5 days of age, the fungal growth inhibiting dilution was taken. The efficacy of the fungicide and humic acid against pathogenic fungi was tested using Poisoned Food Technique, where

the PDA medium was prepared in a glass beaker, then Topsin-M was added to the beaker containing the culture medium and before it solidified in the beakers at an amount of 1 g / liter of the pesticide. The same method was also used with Humic acid with an amount of 2.5 g / liter of acid and shaken in a regular circular motion to ensure homogeneity poured into Petri dishes, The plates were inoculated with a 0.5 cm diameter disc of the fungal growth of the colony of pathogenic fungi taken from the edge of the colony at the age of 5 days. The experiment was carried out with four replications for each treatment, and 4 dishes were left as a comparison treatment, that is, the pathogenic fungi alone. The Petri dishes were placed in the incubator at 24 °C, and after the growth of pathogenic fungi in the control treatment reached the edge of the plate, the percentage of inhibition was calculated according to the equation of Montealegre et al.[11].

$$\text{Inhibition \%} = \frac{\text{The average diameter of control colony} - \text{the average diameter of the treated colony}}{\text{The average diameter of control colony}} \times 100$$

2.3. Evaluation of the Efficiency of Bio Control Elements in Protecting Eggplant Plants from Infection with the fungus S.sclerotiorum in Greenhouses and Plastic Houses and Pots

This experiment was conducted in Babylon province, in the greenhouse of Al-Musayyib Technical College / Al Furat Al Awsat Technical University ,In this experiment, a loam soil and peatmoss in a ratio of 1:2 was used after sterilization using commercial formalin, by preparing a solution consisting of 1:50 formalin / water and (20 ml formalin / liter of water, commercial formalin concentration 40%) the solution was used in a ratio of 3 liter of water / m3 of soil [12]Where the soil was collected in the form of a heap, then covered with a plastic cover of polyethylene (agricultural nylon), it was left for 3-4 days under the sun to provide the necessary heat, then the plastic cover was opened and left for 7 with continuous stirring until the sterilization residuals were removed. The soil was distributed on plastic pots and 7 kg of soil for each pots with a diameter of 24 cm from the top and 22 cm from the bottom and a depth of 18 cm.It was planted with seedlings of the Barcelona eggplant (one plant / pots) at the age of one month. The pots were placed according to Completely randomized design, with four plants for each replicate, and with three replicates for each treatment, including the control treatment without adding pathogenic fungi, with appropriate irrigation and fertilization whenever needed, taking into account not to expose the potting soil to drying out, and after the plant arrived at the age of 45 days, the following treatments were added:

1- Control (without any agent). 2 - T.a. 3 - P. f. 4 - Humic acid 5 - P.c. 6 - T.a. + P. f. 7 - T.a. + H. a. 8 - T.a. + P.c. 9- P. f. + H. a. . 10- P. f. + P.c.11 - S.s. 12- S.s. + T.a. 13- S.s. + P. f.. 14- S.s. + H. a. 15- S.s. + P.c. 16 - S.s. + T.a. + P. f. 17 - S.s. + T.a. + H. a. 18 - S.s. + T. a. + P.c. 19 - S.s. + P. f. + H. a. 20 - S.s. + P. f. + P.c. 21 - S.s. + P.c. + H. a. 22 - S.s. + Topsin-M. 23- P.c. + H. a. 24 - S.s. + T.a. + P. f. + H. a. 25 - S.s. + T.a. + P. f. + P.c. 26 - S.s. + P. fl. + P.c. + H. a. 27 - S.s. + T.a + P. f. + P.c. + H. a. 28 - T.a. + P. f. + H. a. 29 - T. a. + P. f. + P.c.. 30 - P. f. + P.c. + H. a.31 - T.a. + P. f. + P.c. + H. a.

The bacteria *P.fluorescens* was grown on Nutrient broth medium (as in the bacterial contrast experiment). Then the bacterial suspension was added to the soil at average of 25 ml / pots of concentration 6.8 x $^{7}10$ colony-forming units / ml (CFU) / ml). Whereas, Fungal biocontrol agents vaccine represented by *T.asperellum* and *P.commune* was added to the seeds of local millet previously prepared by Dewan method [13] for all treatments that require its addition at a rate of 2 g/kg [14].As for the treatments of the pathogenic fungus *S.sclerotiorum* 5 sclerotia of the pathogenic fungus / plant were added surrounding the stem of the plant with the support of these sclerotia with 0.5 diameter discs of the pathogenic fungus after making a longitudinal wound approximately 1 cm deep on the surface of the stem near the crown area of the eggplant plant and after a week of inoculation With bio control elements. The M-Topsin treatment was conducted at an amount of 1 g / liter of water and was used as a spray on vegetative growth with an interval of 14 days between one spray and another according to the recommendations of the producing company. The first spray was sprayed a week before adding the pathogenic fungus as a preventive measure. The plants were followed up and the plants were watered according to the plant's need, taking into account the control of insect pests, mites and spiders, according to the instructions of the Ministry of Agriculture for growing eggplant in Iraq. The results were taken two months after the addition of the pathogenic fungus inoculum and by

calculating the severity of the plant's vegetative infection with the eggplant stem rot disease, according to the following gradation:
0 = no infection. 1 = surrounding the leg with mildew by less than 1/2. 2 = surrounding the leg with mildew by 1/2 to less than full coverage. 3 = surrounding the leg with mildew completely. 4 = plant death. The severity of infection was calculated according to Mckinny's equation [15] and as follows:

$$\text{infection severity} = \frac{(\text{Number of grade plants } 4\times4+\cdots\ldots+0\times0 \text{ number of grade plants})}{4 \text{ x the total number of plants tested}} \times 100$$

The height, fresh and dry weight of the vegetative and root system were measured.

2.4. Evaluation of the Efficiency of the Previous Bio Control Elements in Protecting Eggplant Plants from Infection with the fungus S.sclerotiorum in the Plastic House

The field experiment was conducted in Babylon province in the plastic house of the College of Technology Al-Musayyib / Al-Furat Al-Awsat Technical University. The soil was plowed and smoothed and leveled. Diammonium phosphate fertilizer was added and then divided into six terraces with a distance of 60 cm and a distance between one terrace and another 80 cm. The experiment was designed according to the Randomized Complete Block Design (RCBD) with three replicates for each treatment and each replicate contains four plants. The pre-prepared seedlings, cultivar Barcelona, about a month old, were planted on the line of one terrace, alternately, and the distance between one plant and another was 40 cm, and after the plant arrived at the age of six weeks and one week before contamination with the pathogenic fungus The previous biological parameters mentioned in plastic house experiment were added. The bacteria *P.fluorescens* was grown on Nutrient broth medium (as in the antagonism experiment) and was added to the soil at an average of 50 ml/plant [16] at a concentration of 6.8×710 CFU/ml), while the bacterial control fungi inoculum represented by With *T.asperellum* and *P.commune* loaded on seeds of local millet previously prepared by Dewan method [13] at an average of 20 g / plant [14] for all treatments that require its addition. The treatment of Topsin-M was coundected at an average of 1 g / L of water and for three sprays with an interval of 14 days between one spray and another, and the first spray was a week before adding the pathogenic fungus. Irrigation, fertilization and control of insect pests, mites and spiders were conducted according to the instructions of the Iraqi Ministry of Agriculture for the cultivation of eggplant in Iraq. The results were taken after 90 days of planting by calculating the percentage of the severity of the eggplant stem rot disease according to the gradient that was used in the canopy experiment.

3. Results and Discussion

3.1. Isolation and Phenotypic Diagnosis of White Mold Disease Caused by the fungus S.sclerotiorum on Eggplant

The results of isolation and diagnosis of fungal growths from sclerotia on PDA culture media showed that the pathogenic fungus *S.sclerotiorum* was obtained, where the fungus was distinguished by a white fungal growth that completely covered the culture medium after 4 days of incubation (Fig. 1 -A) and after 7 days, clusters of yarn appeared. The fungus formed in the form of small white lumps at the edge of dish, which later turned into black sclerotia. The formation of sclerotia was also concentrated at the edge of the dish, and this was due to the physical symptoms of the wall of the dish in which fungi were planted, which stimulated the formation of these sclerotia (Fig. 1 -b). These bodies were distinguished by their different numbers and sizes and were of irregular shapes.The results of the microscopic tests of the mycelium showed that it was divided, translucent, branched, and the infection was characterized by the appearance of symptoms on the leaves, necks and stems of the eggplant plant. Shortly before flowering These results were consistent with the characteristics of the fungus *S.sclerotiorum* that causes white rot on the eggplant plant, which was referred to by several studies [17]. After the incubation period is over, the fungus is identified to the level of the genus and species by observing the sclerotia in addition to the characteristics of the fungal colony and the nature of the mycelium and using the molecular diagnosis of the fungus.

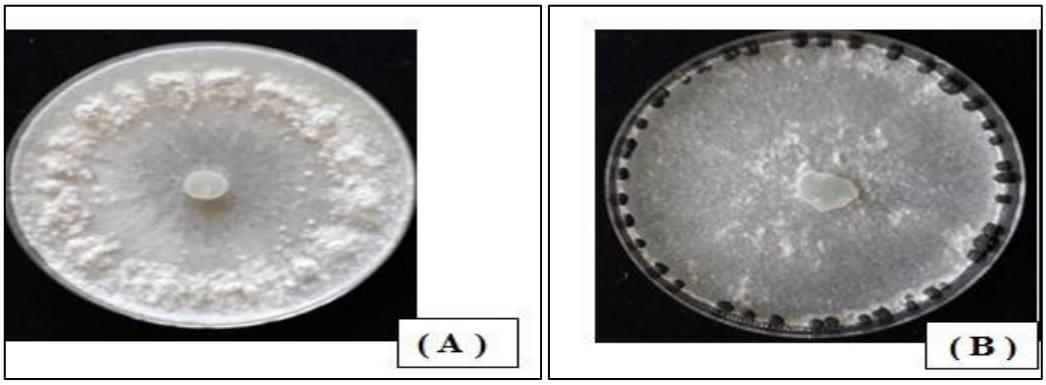

Figure 1. A pathogenic fungus colony developing on the PDA nutrient medium.A- The pathogenic fungus S.sclerotiorum on the PDA medium before the appearance of the sclerotia. B- The pathogenic fungus S.sclerotiorum on the PDA medium after the appearance of the sclerotia.

3.2. Testing the Antagonistic Ability of Some Bio Control Elements Against the fungus S. sclerotiorum in the Laboratory

The results of (Table 1) showed a high antagonistic ability between the bio control elements and the pathogenic fungus *S.sclrerotiorum*, where the treatment of *P.fluorescens* against the pathogenic fungus gave the highest inhibition rate for the pathogenic fungus, reaching 100% compared to the control treatment, which amounted to 0.0%.This is due to the fact that these bacteria have many mechanisms through which they work to inhibit the pathogenic fungus, and one of these mechanisms is the production of antibiotics that are responsible for the inhibition of plant pathogens in general, such as (PLT) Pyuluteorin, (Prn) Pyrolnitrin, (PCA) Carboxylicacid, Phyenazin and (DAPG) Diacetylphloroglycinol-2,4 [18]. The treatment of *P.commune* with the pathogenic fungus gave a high rate of inhibition, which amounted to 86.1% compared to the comparison treatment, which amounted to 0.0%. The reason for the inhibition may be due to the ability of the fungus *Penicillium* spp. On the production of secondary metabolites belonging to the compounds of Naphthalenoids, which have proven effective inhibition against a group of microorganisms [19]or this is due to its production of antibiotics, which is one of the important mechanisms of the fungus in bio control , where it works to produce lovastatin, in addition to its ability to produce oxacillin, statin production, and anti-disease products [20]. This result is consistent with what was found by Al-Jubouri [21], who found that the fungus *Penicillium* sp. It recorded a degree of antagonism of 2 according to the Bell scale (1982) against the pathogenic fungus *S.sclrerotiorum*. Also, the treatment of the fungicide M-Topsin with the pathogenic fungus gave a percentage of 100% inhibition, while the treatment of humic acid did not notice any inhibition of the pathogenic fungus, as the percentage of inhibition was 0.0%.This is due to the humic acid making the medium acidic and suitable for the growth of pathogenic fungi.

Table 1. Test the antagonistic ability of some bio control elements against *S.sclerotiorum* in the laboratory.

No.	Treatments*	Inhibition (%)
1	*T. asperellum*+ Ss	70.00
2	*P. commune* + Ss	86.10
3	*P.fluorescens* + Ss	100
4	Humic acid) (HA + Ss	0.00
5	M - Topsin + Ss	100
6	Ss (control)	0.00
7	L.S.D. (5%)	5.31

Each number in the table represents an average of four replicates. Ss= *S.sclerotiorum**

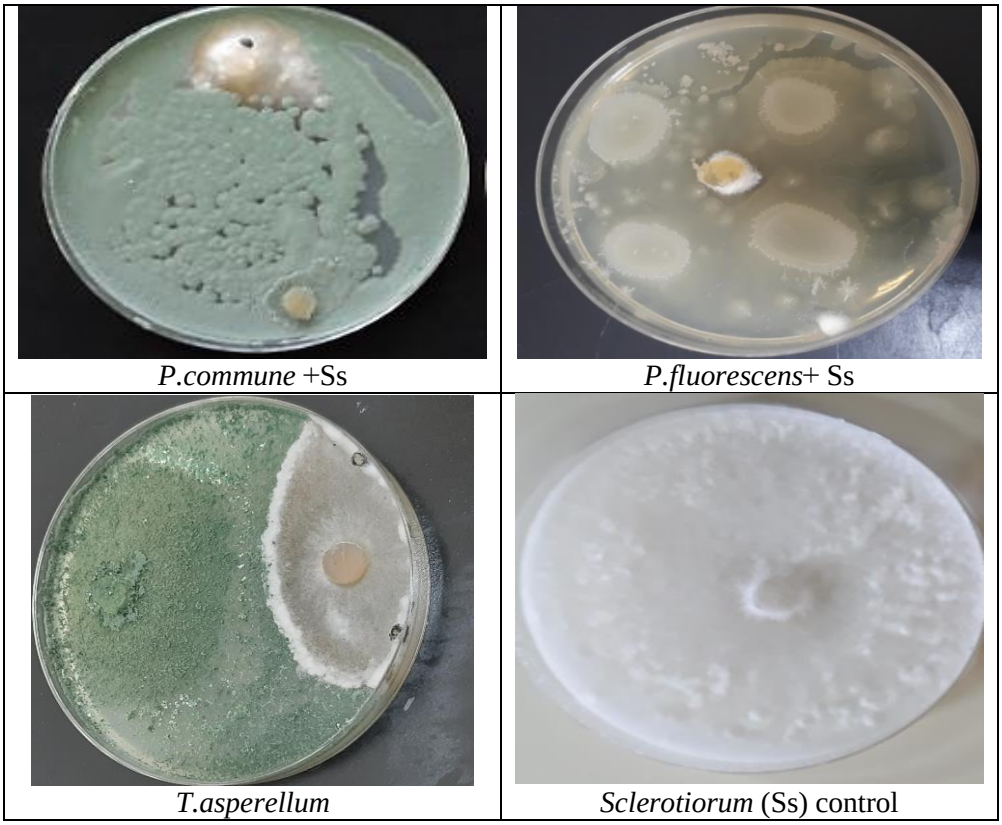

Figure 2. Antagonistic ability of bio control elements against pathogenic fungi *S. Sclerotiorum* (Ss) in addition to the control fungus pathogen alone.

3.3. Evaluation of the Efficiency of Bio Control Elements in Protecting Eggplant from Infection with the Pathogenic fungus S.sclerotiorum in the Greenhouse and Under in the Plastic House Conditions

The results of the plastic house and under the conditions of the greenhouse (Table 2) showed that all treatments used in bio control to white mold disease on the eggplant had a significant reduction in the percentage of infection severity compared to the control treatment (pathogenic fungus alone), the results showed that treatments *T.asperellum* and *T.asperellum* + *P. fluorescens* and *T.asperellum* + *P.commune* and *P.fluorescens* + *P.commune* and *T.asperellum* + *P.commune* + *P. fluorescens* and *T.asperellum* + *P.commune* + *P.fluorescens* + humic acid gave the highest reduction in the percentage of infection severity of pathogenic fungi, reaching 5.55% for all treatments in a row, which was positively reflected on plant growth parameters. Compared to the control treatment in which the percentage of infection severity was 94.44%. It was followed by all of the treatments that included *P.commune*, *P.fluorescens* + *P.commune* + humic acid and *P.fluorescens* in reducing the percentage of infection severity with pathogenic fungi, which reached 8.89, 22.2, 27.78%, respectively. While the pesticide treatment (M-Topsin) and humic acid gave the least reduction in the percentage of infection severity, as it reached 50.00 and 88.33%, respectively and that all the bio treatments used in the experiment (*T.asperellum*, *P.commune* and *P.fluorescens*) gave high significant differences compared to the control treatment (pathogenic fungus alone) through the increase in growth parameters of eggplant plants, it gave asperellum treatment. T+ *P.commune* + *P.fluorescens* in the presence of pathogenic fungi, high results for growth parameters of eggplant plants, Followed by the treatment of fungus. *T.asperellum* + *P.commune* In the presence of the pathogenic fungus, a significant increase in growth parameters such as height, fresh and dry weight of the vegetative and root groups was recorded. This result is in agreement with many studies that showed that the interaction between bio control agent is more effective in reducing the percentage of infection severity with plant pathogens compared to if a biological agent was used alone [22].

Table 2. Evaluation of the efficiency of bio control elements in protecting eggplant plants from infection with the fungus S.sclerotiorum in pots in Babylon province.

No.	Treatments*	infection severity %	vegetative length (cm)	Vegetative weight (g)		Root Total Length (cm)	root total weight (g)	
				Fresh	Dry		Fresh	Dry
1	single plant	0.00	73.11	729.50	139.41	34.16	143.5	47.83
2	T.as	0.00	88.34	1333.75	277.25	49.83	288.08	92.41
3	Pf	0.00	85.11	1025.08	245.08	46.41	280.83	71.33
4	HA	0.00	84.00	1086.25	235.58	51.66	316.58	89.66
5	P.c	0.00	95.00	1455.41	270.41	48.50	278.83	79.25
6	P.f + T.as	0.00	92.66	1152.91	188.16	40.08	232.66	65.16
7	HA + T.as	0.00	87.44	1343.25	235.91	47.83	381.41	96.08
8	P.c + T.as	0.00	96.22	1110.58	257.16	54.00	390.58	95.91
9	HA +P.f	0.00	84.78	1035.91	204.08	42.83	240.83	65.16
10	P.c + P.f	0.00	91.89	1460.08	236.16	40.08	212.33	83.00
11	S.s	94.44	46.66	193.33	112.25	21.16	51.66	16.41
12	T.as + S.s	5.55	83.67	1230.33	193.91	47.33	293.83	78.25
13	P.f + S.s	27.78	70.44	753.41	159.25	38.16	170.66	55.33
14	HA + S.s	88.89	54.11	303.16	121.91	23.00	98.58	25.41
15	P.c + S.s	8.33	81.33	1039.58	186.25	45.25	281.41	71.83
16	Topsin-M S.s	50.00	65.39	569.41	142.16	31.33	139.33	36.16
17	HA + P.c	0.00	101.11	1115.25	238.58	49.83	277.50	86.33
18	P.f + T.as + S.S	5.55	77.89	770.00	163.33	40.00	224.50	62.41
19	HA + T.as + S.s	25.00	76.55	1148.00	186.25	36.58	274.83	83.33
20	P.c + T.as + S.s	5.55	92.89	1273.08	231.58	49.25	345.66	85.25
21	HA + P.f +S.s	50.00	63.22	575.16	140.5	35.25	127.08	40.83
22	P.c + P.f + S.s	5.55	90.89	1172.25	225.5	42.33	315.66	74.08
23	HA + P.c + S.s	22.22	75.44	1118.16	178.25	36.75	182.50	51.41
24	HA + P.f + T.as	0.00	93.45	1353.66	255.25	46.66	302.58	82.66
25	P.c + P.f + T.as	0.00	98.72	1450.41	274.00	54.08	391.66	92.83
26	HA + P.c + P.f	0.00	93.00	1491.75	259.41	45.58	304.41	86.58
27	HA + P.c + P.f + T.as	0.00	96.89	1370.25	272.58	46.75	311.52	89.83
28	HA + P.f + T.as + S.s	8.33	84.55	1049.75	219.50	44.00	266.00	75.25
29	P.c + P.f + T.as + S.s	5.55	96.89	1352.75	227.91	49.75	378.5	86.16
30	HA+ P.c + P.f + S.s	22.2	75.08	1007.33	187.83	38.33	265.17	69.33
31	HA+ P.c +Pf+ T.as +S.s	5.55	88.50	1012.25	206.66	41.58	288.5	77.91
32	LSD%5	17.389	16.463	310.47	71.732	7.3895	124.88	39.118

As for the bio agent treatments that were alone in the presence of pathogenic fungi, *T.asperellum* treatment gave the highest values of growth parameters of eggplant plants compared to the control treatment, followed by the treatment of *P.commune* in giving high efficiency of growth parameters of eggplant plants compared to the control treatment and this is attributed to These fungi are plant growth

promoting fungi. As it has the ability to confer many benefits to the plant by encouraging growth and increasing yield in addition to protecting against pests and pathogens through the degrading enzymes released by these fungi or the biological compounds or antibiotics they produce against plant pathogenic fungi and that the most important fungal species enhancer For root growth (PGPF) is the fungus spp. *Trichoderma* spp. and *Pinicillium* spp. and *Gliocladium* spp. [23], where these types of fungi allow benefiting from decomposing organic matter by dissolving minerals to produce bioactive substances such as plant hormones [6].Recent discoveries indicate the production of large quantities of gibberellins by this fungus (PGPF), which greatly enhances the explanation of the mechanism involved in its effects through the promotion of plant growth as well as the action of *Penicillium* spp. It stimulates and stimulates root growth by another important plant growth hormone, auxin, which plays a major role in promoting plant growth [24]. Also, the treatment of *P.fluorescens* in the presence of the pathogenic fungus gave high results for the growth parameters of eggplant plants compared with the control treatment. These results show that these bacteria have the ability to protect the eggplant plant from infection by pathogenic fungi. This is due to the fact that the use of the PGPR group, which includes *P.fluorescens* bacteria, leads to a significant increase in the quantity of production and a good quality of production, where these bacteria produce Plant growth stimulants such as gibberellins and auxins increase plant growth and productivity [25].The treatment of Topsin-M also gave the least reduction in the severity of infection, due to the rapid spread of the pathogenic fungus and within the tissues of the plant and its carrier vessels. The treatment of humic acid with the pathogenic fungus is also due to the least reduction in the severity of infection due to making the growth medium of the pathogenic fungus acidic and this is in favor of the pathogenic fungus. It ranges between 4 - 5.5, and it was found that the pH of the culture medium is a strong regulator of oxalic acid production, as acid production increases in an acidic environment. Some studies showed that the production of oxalic acid in the fungus *S.sclerotiorum* is stimulated by the enzyme Oxaloacetate Acetylhydrolase, and that the activity of the enzyme increases with increasing pH of the surrounding environment [26].The results also showed a significant increase in the growth parameters of eggplant plants in the treatments that included bio control factors only, whether they were singly or intertwined, compared with the treatment of the plant alone and without adding the pathogenic fungus, it achieved a significant increase in the height, fresh and dry weight of the vegetative and root system.

3.4. Evaluation of Efficiency of Bio Control Agents in Protecting Eggplant from Infection with the fungus S.sclerotiorum in the Greenhouse

The results of (Table 3) of the field experiment in the greenhouse showed that all the bio control treatments used against the white mold disease caused by the fungus S.sclerotiorum on the eggplant caused a significant reduction in the percentage of infection severity and provided good protection for the eggplant, which ranged from 5.55-66.66% compared to with the control treatment (pathogenic fungus alone), which amounted to 83.33%.As the results showed that the transactions. *T.asperellum* + *P.commune* and *T.asperellum* + *P.commune* + *P. fluorescens* gave a high efficiency in reducing the percentage of infection severity of the pathogenic fungus *S.sclerotiorum*, which amounted to 4.16% for both treatments against the pathogenic fungus, followed by treatments. *T.asperellum*, *P.commune*, and *T.asperellum* + *P.fluorescens*, *P.fluorescens* + *P.commune*, *P.commune* + humic acid and *T.asperellum* + *P. fluorescens* + humic acid and *P.commune* + *P. fluorescens* + humic acid and *T.asperellum* + *P.commune* + *P.fluorescens* + humic acid against pathogenic fungus was 8.33% for all treatments, respectively, compared to the control treatment, which gave *T.asperellum* + humic acid, pesticide treatment (Topsin-M), P.fluorescens + humic acid treatment and humic acid treatment lowered the severity of infection, as it reached 16.66, 41.66, 66.66 and 66.66, respectively. Also, all treatments used in bio control achieved high significant differences through the increase in growth parameters of eggplant plants such as height, fresh and dry weight of the vegetative and root group, in addition to the weight of fruits in the presence of the pathogenic fungus *T.asperellum* + *P.commune* + *P. fluorescens* in the presence of the pathogenic fungus *S.sclerotiorum* showed high results for the growth parameters of eggplant plants in comparison with the control treatment in which the height and the fresh and dry weight of the vegetative and root groups in addition to the weight of the fruits reached 145.58, 60.16 cm, 2109.25, 497.83, 298.09 , 86.83, 19885.33 g, respectively. The reason may

be due to the cooperative effect between the bio control agent used in this experiment as they together and in a synergistic manner in stimulating the systemic resistance of plants against pathogens [22].As for the treatments of bio control agent that were individually against the pathogenic fungus *S.sclerotiorum*, the results showed that the treatment of fungus . *T.asperellum* gave the highest results for growth parameters of eggplant plants compared with the control treatment. These results are in agreement with Singh et al. [27] who found that the fungus *T.asperellum*. It provides complete protection against the pathogenic fungus *Fusarium oxysporum*. f. sp. lycopersici . The cause of fusarium wilt disease on tomato and increased plant growth by increasing the availability of phosphorous, the production of indole acetic acid (IAA) and an increase in the activity of the enzyme Protease, These results agreed with Li et al. [28] who used *Trichoderma* spp. In controlling the fungus *S. sclerotiorum* that causes jet flower blight by spraying the suspended fungus on the flowers of the young plant, it achieved an inhibition in the growth of pathogenic fungus and an increase in the production of pods in the plant. It was followed by the treatment of *P.commune*, which gave a high efficiency of growth parameters for eggplant plants compared to the treatment. The ability of *P.commune* to reduce the severity of plant infection with pathogenic fungus and increase plant growth parameters is due to its possession of a number of mechanisms through which it inhibits the growth and pathogenicity of the pathogen, including Its ability to produce many different compounds, and the fungus *P.commune* secretes many encouraging and stimulating substances for plant growth, which is reflected positively on the increase in plant growth indicators of the plant. These results agreed with Al-Jubouri [29], who found that *P.corylophilum* works to resist many pathogenic fungus , including *Fusarium* spp. And *Rhizoctonia solani* and *Macrophomina phaseolina* also noticed a significant increase in the growth parameters of pepper plants in the treatments included in this bio fungus. As for the treatment of *P.fluorescens*, it gave high significant results for the growth parameters of eggplant plants in the presence of pathogenic fungus , compared with the control treatment, where the bacteria *P.fluorescens* plays an important role in providing additional quantities of element N through various mechanisms, including fixing atmospheric nitrogen or analyzing mineral rocks and liberating elements Which helps the roots to grow and deepen into the soil .Hence, increasing its ability to absorb water and nutrients as well as reducing the population density of plant pathogenic microorganisms in the root periphery area [30].

The results also showed a significant increase in the height, fresh and dry weight of vegetative and root systems, in addition to a rise in the fruit weight of eggplant plants in the treatments that included bio control factors and without adding pathogenic fungus in a single or intercropping form

Table 3. Evaluation of the efficiency of bio control elements in protecting eggplant plants from infection with the fungus *S.sclerotiorum* that causes white mold disease in the plastic house in Babylon Governorate.

No	Treatments *	infection severity %	vegetative length (cm)	Vegetative weight (g)		root length (cm)	root total weight (cm)		Eggplant aggregation weight average(g)
				Fresh	Fresh		Fresh	Dry	
1	single plant	0.00	115.58	1168.75	229.33	42.00	177.16	37.16	8309.00
2	T.as	0.00	139.5	2255.16	524.33	54.58	323.66	74.5	16618.66
3	Pf	0.00	130.5	2153.33	465.25	53.50	281.08	86.25	16078.33
4	HA	0.00	137.5	2125.66	556.16	53.58	304.75	91.91	11641.33
5	P.c	0.00	141.25	2144.16	498.58	54.39	332.41	103.25	17813.25
6	P.f + T.as	0.00	133.66	2272.58	451.58	54.916	237.16	84.66	16289.66

7	HA + T.as	0.00	146.33	2136.41	536.41	55.75	309.37	93.08	17476.66
8	P.c + T.as	0.00	151.33	2285.25	504.00	64.39	308.58	92.916	18950.00
9	HA +P.f	0.00	126.41	1768.00	518.75	50.16	290.00	85.50	15229.00
10	P.c + P.f	0.00	139.33	2175.83	532.83	58.41	356.25	106.75	18134.66
11	S.s	83.33	42.41	641.416	82.75	17.58	47.58	12.08	2570.33
12	T.as + S.s	8.33	135.5	2091.66	483.75	54.16	296.25	81.41	16128.00
13	P.f + S.s	12.5	123.91	1589.75	388.16	48.75	246.08	63.08	15822.00
14	HA + S.s	66.66	62.25	776.916	105.25	23.25	89.00	19.50	5448.00
15	P.c + S.s	8.33	135.00	1915.50	444.66	53.75	222.16	73.41	15697.33
16	Topsin-M +S.s	41.66	93.66	1455.75	180.75	29.91	121.25	33.31	5737.21
17	HA + P.c	0.00	143.91	1952.5	512.50	55.16	275.83	80.62	15862.66
18	P.f + T.as + S.S	8.33	125.00	1832.66	451.33	52.50	259.91	63.42	15016.66
19	HA + T.as + S.s	16.66	144.04	1870.66	460.166	52.16	261.08	62.41	14695.33
20	P.c + T.as + S.s	4.16	144.168	206708.	468.25	58.58	264.50	81.83	18669.00
21	HA + P.f +S.s	66.66	86.91	1025.00	208.00	39.00	79.08	36.16	3610.91
22	P.c + P.f + S.s	8.33	138.05	1829.58	408.66	55.41	274.50	80.25	16313.33
23	HA + P.c + S.s	8.33	137.50	1732.91	444.16	47.75	246.87	58.91	12924.33
24	HA + P.f + T.as	0.00	141.41	1758.41	417.83	57.91	311.12	91.25	18481.66
25	P.c + P.f + T.as	0.00	154.91	2275.50	527.33	66.58	313.00	109.16	20288.93
26	HA + P.c + P.f	0.00	141.00	2015.25	514.08	55.75	312.94	94.58	16844.33
27	HA + P.c + P.f + T.as	0.00	147.00	2395.16	531.91	64.25	270.91	79.08	18145.00
28	HA + P.f + T.as + S.s	8.33	129.41	1806.66	363.91	49.66	234.33	70.25	15654.66
29	P.c + P.f + T.as + S.s	4.16	145.58	2109.25	497.83	60.16	298.09	86.83	19885.33
30	HA+ P.c + P.f + S.s	8.33	138.58	1888.5	417.16	53.00	209.08	64.66	15555.5
31	HA+ P.c +Pf+ T.as +S.s	8.33	142.91	1985.16	455.08	55.16	240.50	74.25	15769.43
32	L.S.D%5	15.108	17.909	625.80	110.21	10.175	139.9	35.877	3711.50

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