Stimulating the defenses of tomato plants against the pathogenic fungus *Alternaria aternata* by the effect of *Tricoderma* species and vitamin B2

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Abstract

This study demonstrated the efficacy of riboflavin and *Trichoderma* spp. In increasing the induction of resistance in tomato plant against the pathogenic fungus *Alternaria alternata* that causes Alternaria spot disease. The study in Invetro showed the ability of two isolates of the pathogenic fungus *A.alternata* in the water agar medium, as the isolate of fruits had the most impact on the severity of infection of tomato seeds, which amounted to 59% compared to isolate the leaves, which amounted to 47%. The results of the antagonism test for *T.viride* and *T.koningii* in PDA medium showed a high antagonistic ability against *A.alternata* by double culture method, where the antagonism degree was 1 according to the Bell scale. Many chemical compounds were obtained from the filtrate of *T.viride* and *T.koningii* by GC-MS technique believed to have a role in inhibiting pathogens, including 2-Propenoic acid and Octadecanoic acid. The bio-resistant fungi and riboflavin used reduced the severity of infection with the pathogen. *A. alternata* in pots had the lowest severity of infection with riboflavin treatment (8.21)%. The highest content of peroxidase enzyme in tomato leaves when *T.v*+*T.k*+B2+*A.a* treatment was 3.30 units/gm.

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Introduction

Tomato (*Solanum lycopersicum* L.), one of the important vegetable crops for human consummation, it constitutes a high nutritional value. It is rich in vitamins, including A, B, and C. It is also characterized by containing many mineral elements such as iron and phosphorous, as well as containing antioxidants, including lycopene (Christoppher et al., 2010 and Rakha et al., 2011).In all its cultivation areas, the tomato plant is exposed to many agricultural pests, the most important of which are fungi, bacteria, nematodes and viruses. The fungus *Alternaria alternata* is among the important causes that affect the tomato plant, as it causes many symptoms, including spots on the leaves, necrosis of the stem, black mold on the fruits, and the infection with the fungus *A. alternata* develop during harvest and after harvest (Tilgen and Geoips, 1982; Matrood and Rhouma, 2021a).Many chemical fungicides have been used to combat leaf spot diseases caused by the fungus *A. alternata* (Matrood and Rhouma, 2021b). Despite the contribution of chemical pesticides to the protection of agricultural production, there are many negative aspects to them, including their environmental risks and their impact on many non-target organisms and the emergence of strains resistant to the action of pesticides (Altman and Campbell, 1977; Matrood and Rhouma, 2021a). Sequeira (1983) also found that the induction of systemic resistance in the plant is accompanied by the accumulation of proteins and other compounds. The isolates of the biogenic fungus *Trichoderma* spp. It has the ability to induce systemic resistance by increasing the activity of peroxidase enzyme in plants treated with the fungus (Hamid, 2002; Matrood and Rhouma, 2021c). Agrious, 1997 noted that the plant defenses are stimulated when exposed to external causes, living or non-living, by the production of phytoalexins as well as their production of suberin and lignin. In addition to the non-living factors where vitamins appear as organic compounds used as a catalyst in the systemic resistance induced in the plant (Reignault and Walters, 2007). Where they are considered effective elements against many pathogens through foliar spraying of different

vitamins alone or in combination with other compounds such as vitamin.

Materials and Methods

Isolation and identification of *A. alternata*

Infected tomato fruits in the form of a spot of olive-black color were brought from the local markets, and the leaves appear spots with yellow color were located from the greenhouse in the Basrah Research Station. The samples were washed with water, sterilized with a solution of sodium hypochlorite NaOCl (10%) for 3 minutes and washed with sterilized. The vegetable samples were placed on the Whatman No.1 filter paper to dry. 2-3 small samples were transferred to Petri dishes containing sterile PDA medium and incubated at 25±2°C for 7 days with three replicates (Matrood et al., 2022).

Pathogenicity test

The two isolates of *A. alternata* were grown on W.A medium and incubated at 25±2 temperature for 3 days, then sterilized tomato seeds were sown with a solution of sodium hypochlorite (NaOCl) at a concentration of 10% for 3 minutes and washed with sterilized distilled water and distributed circularly around the fungal colonies. The experiment were composed with inoculated seeds and control (treated with sterilized distilled) with 10 seeds in each dish. The dishes were incubated at a temperature of 25 ± 2°C for 10 days. The pathogenicity of *A. alternata* was calculated according to a scale from 0 to 5: 0 seeds are intact; 1 Part of the seedlings are colored brown with their connection to the fungus; 2 The fungus invades the seed coat but the seedlings are intact; 3 The seed coat is free from the fungus but infected; 4 Seed coat and seedling infected; 5 The seeds are infected and not germinated (Manici et al., 1992).

Antagonism test between a living fungus and the pathogenic fungus *A. alternata*

The direct confrontation method was adopted, Daul-culture Technique, to test the biological ability of fungi against the pathogenic fungus *A. alternata*. Divide a petri dish containing a sterile PDA medium with an imaginary line into two equal parts and inoculate the center of the first section with a 0.5 cm diameter disc from the colony of the live fungus *T. viride* at the age of 7 days and inoculate the second section of A plate with a diameter of 0.5 cm from a colony of pathogenic fungus *A. alternata* at the age of 7 days, with 3 replications, was used as control. A petri dish containing sterile PDA medium was inoculated with a disk with a diameter of 0.5 from a colony of pathogenic fungus *A. alternata* was incubated at a temperature of 25±2 and upon arrival of the pathogen in the control treatment (Matrood et al., 2021). To the edge of the plate, the degree of antagonism was calculated according to the five-degree scale of Bell et al. (1982).

1. The bio-resistant fungus covers the whole dish
2- The biological resistance fungus covers one third of the plate
3- The biological resistance fungus covers half of the plate
4- The pathogenic fungus covers two-thirds of the plate
5- The pathogenic fungus covers the whole plate

Compounds present in filtrate of bio-resistant fungi and their identification by GCMS technology

The filtrate of the two fungi prepared in paragraph 3-5 was taken. Then the filtrate of each *T. viride* and *T. koningii* was placed in Petri dishes and placed in the freezer for one day after freezing. It was transferred to the Freeze dryer. Then the samples were extracted using 10% ethanol, then 0.2 ml of the extract was taken. It was injected with a GCMS device to read the substances and compounds present in the filtrate (Senthikumar et al., 2011).

Study of the biological effect of bio-resistant fungi and riboflavin in stimulating and encouraging plant growth in plastic pots

The experiment was carried out in pots of dimensions 17 x 18 cm. Mixed soil was sterilized mixed with moss at a ratio of 1:2 ratio. The experiment design was in BAC and composed with plants treated with *T. viride* and *T. koningii* in the presence of the pathogen. with three seedlings in each pot, at the beginning of the plants reaching the flowering stage, they were sprayed with riboflavin solution at a concentration of 15 mmol/mol, and the pesticide treatments were also sprayed with a concentration of 50 ppm, then after three days they were sprayed with a suspension of the

pathogenic fungus *A. alternata* spores (Matrood and Rhouma, 2022). After a month, the disease severity was calculated according to Mckinney's (1923) (Al-Waeli, 1988).

The content of peroxidase enzyme (Matrood and Rhouma, 2021a).

They were transferred from the field to the laboratory inside a box containing ice, then 1 gm of roots was taken, then washed with deionized distilled water and mashed in a ceramic mortar inside an ice bath. bath) by adding 2.5 ml of Potassium phosphate buffer with a concentration of 0.05 mo with a pH of 6 = pH was prepared by dissolving 6.804 g of potassium dihydrogen phosphate (KH2PO4) + 8.709 g of potassium phosphate (K2HPO4) in 100 ml of distilled water. , take 5 ml of the mixture and complete the volume to 100 ml of distilled water to obtain a phosphate buffer solution at a concentration of 0.05 molarity 6 = pH. The mixture (roots + phosphate buffer solution) was placed in a centrifuge at 1200 rpm for 20 minutes, then 250 microliters were added From each dye Gauiacol at a concentration of 0.5%, hydrogen peroxide at a concentration of 0.3% v/v and 2.5 of a phosphate buffer solution, then the readings were recorded by reading the amount of optical absorption of the sample in a spectrophotometer at a wavelength of 470 nm after Filter the device with a mixture consisting (5 ml of buffer solution and 250 µl each of Gauiacol dye and hydrogen peroxide after combining them, then 3 ml was taken and placed in the device cell for filtering). grams of soft weight according to the following equation:

Enzymatic activity Absorption unit/g = (the device read) / (reading taken volume x (form weight) / (extraction volume) x 100 (Matrood, 2015)

Results and Discussion

Isolation and identification of *A. alternata*

Isolation of the fungus *A. alternata* from tomato leaves and fruits (Fig. 1), which appeared on dark brown spots tilted to black. The spots are characterized by being of different shapes and sizes and are often surrounded by a yellow halo. The fungus colonies were distinguished by the fact that their upper surface was olive-black with a texture Velvety and irregular in shape, the surface of the lower colony is brownish-black, and the colonies were slow-growing on the medium of PDA, and when examined under a microscope at a power (40x) the spore carrier appeared as a single bearing a series of small-sized spores characterized by the presence of three to eight transverse septa with the presence of Longitudinal septums are often neckless or short neck sporangia and these characteristics are consistent with Chen (2018), Alubi (2017) and Matroud (2009).

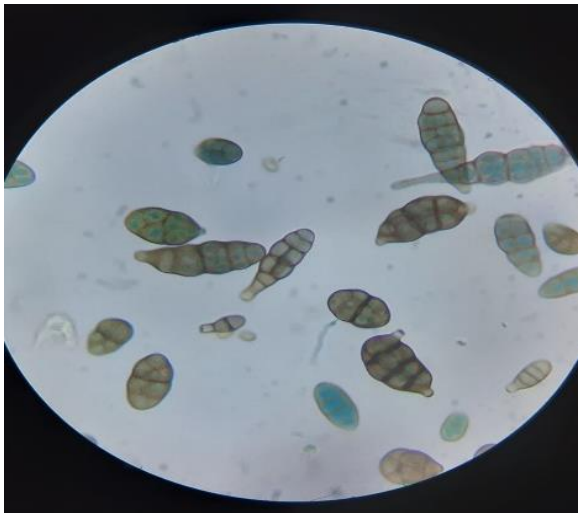


Figure (1) Isolation and diagnosis of the pathogenic fungus *A.alternata*

Pathogenicity test

After obtaining the two isolates of the pathogenic fungus *A. alternata*, isolate was given number 1 isolated from tomato fruits and isolate was given number 2 isolated from leaves, and when testing the pathogenicity of the two isolates in the middle W.A. that isolate number 1 was more effective in the percentage of seed infestation, as it reached. The disease severity of the first isolate were highly (59%) compared to the isolate from leaves (47%) (Table 1).

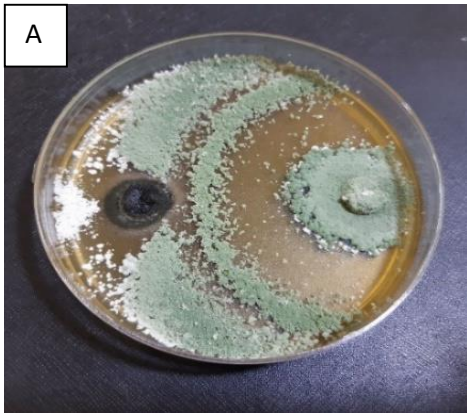
Table (1) The percentage of disease severity of two isolates of pathogenic fungus *A.alternata* in medium W.A with tomato seeds.

A. alternata	disease severity
Isolate No.1	59
Isolate No.2	47
L.S.D 0.01	8

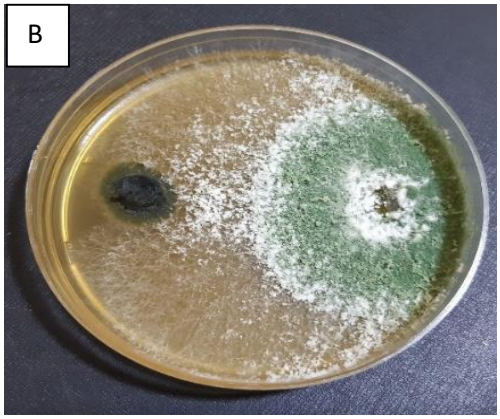
Evaluation of the biological efficiency of fungi in the growth of pathogenic fungi *A. alternata*

The results of the double culture experiment showed the ability of *T.viride* to inhibit the growth of *A.alternata* in the PDA culture medium, where the degree of antagonism was 1 according to the scale of Bell et al. (1982), if the fungus *T.viride* covered the whole plate compared to the fungus The pathogen.

Inhibiting the pathogenic fungus is due to the rapid growth of *T.viride*, competition for nutrients, and the ability to produce cell wall-dissolving enzymes that work within the mechanisms of antagonism and parasitism to break down the cell wall of pathogenic fungi, including Chitinase, Cellulase, Glyconase B-1,3- and Protenase (De Marco and Felix, 2002 & Howell, 2003). In the middle is the PDA. As for the results of the antagonism by the double culture method of *T.koningii* in inhibiting the pathogenic fungus *A.alternata* in PDA culture media, it reached a degree of antagonism of 1 according to the scale of Bell et al., (1982). If the fungus covers the whole plate compared to the pathogenic fungus, Melo and Faull (2000) showed that three isolates of *T.koningii* and *T.harzianum*inhibited the growth of *Rhizoctoniasolani*in PDA culture media. The reason for the inhibition may be due to the secretion of metabolites that contain inhibitory toxins of the pathogen.Rajkonda et al. (2011) indicated the ability of species of *Trichoderma*spp, represented *by T. koningii, T.viride*, *T.harzianum*, *T.virens* and *T.psedokoningii* to inhibit pathogenic fungi in the food media, while they differed in their ability to inhibit pathogenic fungi.



A: T.koningii +A.alternata



B:T.viride +A.alternata

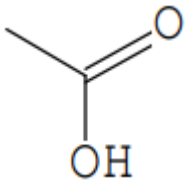
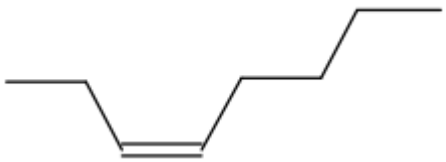
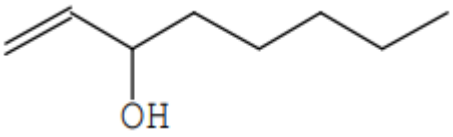
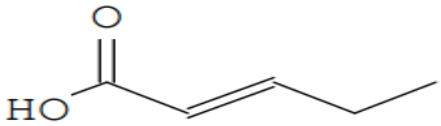
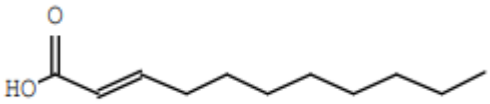
Identification of compounds present in the filters of *T.viride* and *T.koningii* by GC-MS

According to the results, a table (2) was obtained on many compounds and substances present in the filter of the fungus *T.viride* and *T.koningii*, some species of the genus *Trichoderma* are used as biological control factors due to their possession of many mechanisms, including fungal parasitism, antagonism, competition, enzymatic activity and role in induction of defense mechanisms In the plant in addition to its production of many active volatile organic compounds (Sood et al., 2020). In previous studies, it was found that VOCs from microorganisms have the ability to stimulate genes responsible for the defense mechanism in plants and also prevent infection caused by pathogens (Cordovez et al., 2017).

Trichoderma species has significant activity against many plant pathogenic fungi including the pathogenic fungus *Fusarium* (Zhang et al., 2014). Additionally, it has been extensively studied for its effective role as biofertilizer and in pest management (Gupta et al., 2014).

These compounds have been found to promote plant growth by protecting it from pathogen attack (Schulz-Bohm et al., 2017 and Tilocca et al., 2020). The volatile compounds produced by *Trichoderma* species interact between plants and organisms. These produced compounds enhance plant biomass and also compete with the growth of pathogens, as they act in a complex mixture. Environmental conditions such as humidity, temperature, etc. affect production and mechanism of action (Tilocca et al., 2020).

Table (2) Compounds in the filtrate of *T.viride* and *T.koningii* with GC-MS . technology

chemical compound name	chemical composition	chemical formula	molecular weight
Acetic acid		C2H4O2	60
3-Octene		C8H16	112
1-Octen-3-ol		C8H16O	128
2-Pentenoic acid		C5H8O2	100
2-Nonenal		C9H16O	140
2-Undecenoic acid		C11H20O2	184
Octadecanoic acid, docosyl ester		C40H80O2	592

Effect of provoking factors on the severity of tomato pathogenic fungus infection in the pots

The results as shown in Table (3) that inducing factors and biological resistance fungi have good efficiency in reducing the severity of infection in tomato plants with the pathogenic fungus *A.alternata* The infection severity decreased from 53.57% in the pathogenic fungus treatment to 8.21, 8.33 and 16.66% in the riboflavin treatment and the interaction treatment. *T.v* + *T.k* +B2 + *A.a* and the fungus *T.k*+*A.a* treatment respectively.

The effects of biological fungicides in reducing the reduction of pathogenic fungi in tomato plants can be explained by the efficiency of biological resistance factors *Trichoderma* spp. In reducing plant infestation with the pathogen, which is due to the increase in the density of the population in the soil and attacking the pathogens and destroying the reproductive parts of other fungi (Lewis and Lumsden, 2001), Kucuk and Kivanc (2003) also confirmed that the addition of the biological fungus *Trichoderma* to the soil led to the inhibition of the fungus *Sclerotiumrolfsii* and *R.solani* by 82.8% and 76.6%, respectively. The pesticide

treatment reduced the effect of the pathogenic fungus, the infection severity was 33.33% compared to the pathogenic fungus treatment, which reached 53.57%. The cell in the pathogenic fungus, which causes a defect in the growth of the fungus and thus leads to its death (Wang et al., 2015).

Table (3) The role of riboflavin and fungi in the severity of infection with A.alternata fungus in pots

Treatment	Injury severity %
A.alternata	53.57
Ortiva *+A.a*	33.33
B2*+ A.alternata*	8.21
T.viride+ A.a	25.02
T.koningii+ A.a	16.66
*T.v+T.k+*B2*+A.a*	8.33
LSD 0.05	6.1

Determination of peroxidase enzyme in roots and leaves of tomato

The results of the experiment showed Table (4) when plants were treated with riboflavin (B2) and biological resistance factors, whether the plants were inoculated with the pathogenic fungus or not, led to an increase in the activity of the peroxidase enzyme, whether in the roots or leaves, where the interaction treatment *T.v* + *T.k* + B2 + *A.a* reached the highest concentration The enzymatic activity is 3.30 units/gm fresh weight of the shoot, this may be due to the ability of riboflavin and biological resistance factors to stimulate peroxidase enzyme and increase its activity in addition to the enzymes secreted by biological fungi.

The role of the peroxidase enzyme in inducing resistance in plants through its ability to synthesize phytoaloxins inside the plant, during the oxidation of phenols and converting them into quinones that are more toxic than the original compound of the pathogen, and also works on the synthesis of lignin and suberin, which are physical obstacles against the pathogen of the plant (Barcelo et al., 1996).).

Table (4) The content of peroxidase enzyme in the roots and leaves of the tomato plant

Treatment	Enzyme estimate unit/gm
Control	1.66
Ortiva	1.33
T.V	1.71
T.K	0.86
A.a	1.86
B2	2.08
T.V+A.a	1.65
T.K+A.a	1.28
B2+A.a	2.18
*A.a+*Ortiv*a*	0.93
T.V+T.K	1.93
T.K+B2	2.18
T.V+B2	1.91
T.V+T.K+B2	1.08
T.V+T.K+B2+A.	3.30
LSD 0.05	0.45

References

Agrios, G.N. (1997). Plant Pathology. 4th Ed. Academic Press Inc. New York. P 635.

Altman , J . and Campbell . (1977). Effect of herbicides on plant disease Annual Review of Phytopatology . 15: 361- 385

Al-Uqbi, Zahraa Abdul-Latif Jassim ((2017). Evaluation of the efficacy of the biological preparation of *Trichoderma viride* and organic fertilizers in combating *fusarium* wilt and tomato leaf spot. Master's thesis / University of Basra. 84 pg.

Al-Waeli, Dia Salem Ali (1988). Studies on dependent blight on tomato caused by the fungus (ELL). Grout & Jones *Alternaria solani* Master Thesis, College of Agriculture / University of Baghdad. 96 pages

Barcelo, A.R; Zapata, J.M. and calderon, A.A. (1996). A basic peroxidase isoenzyme , marker

of resistance against Plasmoparaviticola in Grapevines, is induced by an Elicitor from *Trichoderma viride* in susceptible. Phytopathology. 144: 309-313.

Bell,D,K;Wells,H.D and Mirkham,C.R.(1982).Inveitro antagonism of *Trichoderma* species angainsts six fungal plant pathogen .phytopathology.72:379_382.

Chen,Y.J;Meng,Q;Zeng,L.,andTong,H. R(2018).Phylogenetic and morohological characteristics of *Alternariaalternata* causing leaf spot disease oncamelliasinensis in China.Australasian Plant Pathology,47(3),335-342.

Christopher,D.J.,T.S.Raj,S.U.Rani and R.Udhyakumar.(2010).Role of defense enzymes activity in tomato as induced by *Trichoderma virens* against Fuzarium wilt caused by *Fusarium oxysporium*f.sp.lycopersici.Journol of Biopesticides 3(I special Issue) 158-162.

Cordovez,L.R.Pierik,R.Mumw,V.J.Carrion,J.M.Raaijmakers.(2017)Plant phenotypic and transcriptional changes induced by volailes from the fungal root pathogen *Rhizoctonia solani*.Fort.Plant Sci.,8,p.1262

De Marco, J. and Felix, C. R. (2002). Characterization of aproterse produced by aTrichdermaharizanum isolate which controls Cocoa plant witches broom disease. BMC Biochemistry 3 : 1 -7.

Gupta.M.Schmoll,A.HerreraEstrella,R.S.Upadhyay,I.Druzhinina,M.Tuohy.(2014) Biotechnology and biology of *Trichoderma*Newnes .

Hamid, Fakher Rahim. (2002) Study of the efficacy of isolates of the fungus *Trichoderma*.spp. In inducing resistance against *Rhizoctonia solani* and stimulating growth in four cotton cultivars. Master Thesis, College of Agriculture, University of Baghdad

Howell, C. R. (2003). Mechanisms employed by *Trichoderma* species in the biological control of plant disease : the history and evolution of current concepts. Plant Dis., 87, 4-10.

Kucuk, C. and Kuvane, M. (2003). Isolation of *Trichoderma* spp. and determination of their antifungal, Biochemical and physiological features. Turk. Journal Biol. 27: 247-253pp

Lewis, J.A. and R.D. Lumsden. 2001. Biocontrol of damping-off of greenhouse- grown crops caused by *Rhizoctoniasolani* and saprophytic growth of the pathogen in soilless mix. Plant Dis. 82: 501-506.

Manici, L. M., Cerato, C., and Caputo, F. (1992). Pathogenic and biological variability of Macrophominaphaseolina (Tassi) Goid. isolates in different areas of sunflower cultivation in Italy. 13th Proc. of Intern. Sunflower Conf, 1, 779 – 784.

Matrood A.A.A, Rhouma A. and Mohammed T.F., (2022). Control of Fusarium wilt disease of cucumber using rhizospheric antagonistic fungi. Arab Journal of Plant Protection, 40(1): 62-69. https://doi.org/10.22268/AJPP-040.1.062069

Matrood A.A.A. and Rhouma A., (2021a). Evaluating eco-friendly botanicals as alternatives to synthetic fungicides against the causal agent of early blight of Solanum melongena. Journal of Plant Diseases and Protection 128, 1517-1530. https://doi.org/10.1007/s41348-021-00530-2

Matrood A.A.A. and Rhouma A., (2021b). Efficacy of foliar fungicides on controlling early blight disease of Eggplant, under laboratory and greenhouse conditions. Novel Research in Microbiology Journal, 5(3): 1283-1293. https://doi.org/10.21608/NRMJ.2021.178310

Matrood A.A.A. and Rhouma A., (2021c). Potential of the indirect and direct beneficial effects of the use of *Trichoderma koningii*, *Aspergillus niger* and *Mucor* sp. on eggplants plants: Plant growth and systemic resistance induction. Nova Biotechnologica et Chimica, 20(1): e809. https://doi.org/10.36547/nbc.809

Matrood A.A.A. and Rhouma A., (2022). Bioprotection of *Cucumis melo* from Alternaria leaf spot by *Glomus mosseae* and *Trichoderma harzianum*. Tropicultura, 40(2): 1-11. https://doi.org/10.25518/2295-8010.2075

Matrood A.A.A., Rhouma A. and Okon G.O., (2021). Evaluation of the biological control agent's efficiency against the causal agent of early blight of *Solanum melongena*. Arab Journal of Plant Protection, 39(3): 204-209. https://doi.org/10.22268/AJPP-039.3.204209

Matroud, Abd al-Nabi Abd al-Amir (2009). Effect of some insecticides on tomato leaf spot disease caused by Fr (keissler) *Alternaria alternata*. Master Thesis. College of Agriculture / University of Basra. 54 pages

Matroud, Abd al-Nabi Abd al-Amir (2015). Integration in the control of charcoal rot disease caused by the fungus *Macrophomina phaseolina* (Tassi) Goid PhD thesis. College of Agriculture / University of Kufa. 154 pages

Melo, I. S. D. and Faull, J. L. (2000) . Parasitism of *Rhizoctonia solani* by strains of *Trichoderma* spp. Scientia Agricola, 57(1): 55-59

Rajkonda, J. N.; Sawant, V. S.; Ambuse , M. G.andBhale, U. N. (2011) . Inimical potential of *Trichoderma* species against pathogenic fungi . Journal Plant Sciences Feed, (1) : 10- 13.

Rakha, M.,J.Scott and S. Hutton. (2011) Identification of ichomes, loci and chemical compounds derived from Solanumhabrochaites accession LA1777 that are associated with resistance to the sweet potato whitefly, Bemisiatabaci in tomato, *Solanumlycopersicum*. 43rd Tomato Breeders Roundtable Meeting. University of Florida. pp.31.

Reignault P, Walters D.(2007) Topical application of inducers for disease control. In: Walters D, Newton A, Lyon G, editors. Induced resistance for plant defence: a substainable approach to crop protection. Oxford, UK: Blackwell Publishing;. p. 179e200.

Schulz-Bohm, K., Martı´n-Sa´nchez, L., Garbeva, P.,(2017). Microbial volatiles: small molecules with an important role in intra-and interkingdom interactions. Front. Microbiol. 8, 2484.

Senthilkumar, G.Madhanraj.P. Panneerselvam.(2011). Studies on the Compounds and Its Antifungal Potentiality of Fungi Isolated From Paddy Field Soils of Jenbagapuram Village, Thanjavur District, and South India. Asian J. Pharm. Res. Vol. 1: Issue 1, Pg 19-21.

Sequeira, L. (1983). Mechanisms of induced resistance in plant. Ann. Rev. Microbiology. 37:51-79.

Sood,D Kapoor ,V .Kumar, M.S. Sheteiwy ,M .Ramakrishnan ,M. Landi,F .Araniti ,A.Sharma(2020).Trichoderma:The secrets of amultitalented biocontrol agent plant,9,p.762.

Tilgen,W.H.and Gepois.(1982) Nature and etont of losses in fresh kepaked Tomato. phytopathology .72:266_267.

Tilocca, B., Cao, A., Migheli, Q., (2020). Scent of a Killer: microbial volatilome and its role in the biological control of plant pathogens. Front. Microbiol., 116:25-28. 635 pp.

Wang, L; Zhu, X; Liu, J; Chu, X; Jiao, J; Liang, Y (2013) Involvement of phospholipases C and D in the defense responses of riboflavin-treated tobacco cells. Protoplasma 250, 441-449.

Zhang,X.Yang,W.Ran, Q. Shen. (2014) .*Fusarium oxsporum* induces the production of proteins and volatile organic compounds by *Trichoderma harzianum* T-E5.FEMS Microbiol.Lett.,359,pp.116-123.

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