

EFFECT OF *TRICHODERMA HARZIANUM* AND ENDOMYCORRHIZAE ON GROWTH AND FUSARIUM WILT OF TOMATO AND EGGPLANT.

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ABSTRACT

The effect of endomycorrhizae (AMF) and strain of *Trichoderma harzianum* Tcomp was tested on the growth of tomato and eggplant plants against Fusarium wilt caused by the strain FA5 of *Fusarium oxysporum* f.sp. *albedinis* isolated from the date-palm plants attacked by bayoud. The agronomic parameters of tomato plants co-inoculated with AMF and Tcomp are greater compared to other treatments, namely, number of flowers (8), fruits number (1.5), leaves number

(14.5), aerial part length (46 cm), root length (18 cm), fresh weight of the aerial part (38.5 g) and fresh weight of root part (15.1 g). Similarly, eggplant plants had an increase in agronomic parameters following this double inoculation, flowers number (10.3), fruits number (0.5) number of leaves (26 , 5), the length of the aerial part (40.1 cm), root part length (17.3 cm), aerial part fresh weight (42.1 g) and root fresh weight (18.3 g). Tomato plants inoculated with only *F. oxysporum* f. sp. *albedinis* FA5 showed the highest dwarfing indices and leaf alteration indices (DI: 80%; LAI: 0.812). These indices decreased sharply in the application of mycorrhizae only (DI: 2.23%; LAI: 0.06) or *T. harzianum* alone (DI:

3.87%; LAI: 0.065), while the double inoculation *T. harzianum* and mycorrhizae has better inhibited the leaves alteration (LAI: 0,021). Similarly, the co-inoculation of eggplant seedlings with mycorrhizae and *T. harzianum* has generated the weakest DI and LAI (DI: 3.45%; LAI: 0.0816), whereas the controls plants inoculated only with the pathogen have shown the highest indices (IR: 56%; IAF: 0.689). Indeed, co-inoculation with Tcomp and AMF has prevented the migration of *F. oxysporum* f. sp. *albedinis* (FA5) to the aerial part of the two plant species, and has decreased its presence in the root part (Tomato: 3%; eggplant: 6%) It should be noted that the AMF did not hinder the roots colonization with *T. harzianum* Tcomp. Co-inoculation of AMF, with *F. oxysporum* f. sp. *albedinis* or TComp or both at once has not prevented the formation of endomycorrhizal structures, the intensity and spore's number are respectively (63%, 260 spores); (56%, 220 spores) and (48%, 180 spores) for tomato and (45%, 280 spores); (38%, 246 spores) and (36%, 190 spores) for eggplant.

KEYWORDS: Endomycorrhizae, AMF, *Trichoderma harzianum*, *F. oxysporum* f. sp. *albedinis*, tomato, eggplant.

INTRODUCTION

Tomato (*Lycopersicon esculentum* L.) has a high nutritional value, it is an important source of vitamin A and C (Islam and Faruq, 2012). Its water content varies between 93 and 95% (Shankara *et al.*, 2005). The eggplant is grown in many countries due to its high content of bioactive compounds, including phenolic compounds, macro and micronutrients (Luthria, 2009). Both Solanaceae are among 40 vegetable species most produced in the world (FAO, 2008; Aurore Lebeau, 2012) and are grown in all climates, especially in the tropical zones, where their production is grip of many biotic constraints, particularly bacterial wilt (Aurore Lebeau, 2012), powder mildew in eggplant and tomato mildew caused by the fungus *Oidium lycopersici* in the Netherlands (Huang *et al.*, 2000).

Chemical treatments against soil-borne root pathogens are very dangerous; they cause technical environmental and economic problems. These limits of chemical control and the high concern for the preservation of the environment (Alabouvette *et al.*, 2006) are the major reasons for the increased interest in the use of biological control through its ability to provide an effective protection in the long term and without negative impact on the environment or on human health (Demir *et al.*, 2015).

Among the biological control agents commonly used, the mycorrhizal fungi (AMF) are symbiotic microorganisms present in the rhizosphere. They bring considerable benefits to plants in terms of resistance and Development (Azcón-Aguilar and Barea, 1996; Smith and Read, 2008).

Similarly, *Trichoderma* species are often successfully used as biological control agents against various pathogenic fungi (Monte, 2001, Paulitz and Belanger, 2001). The objective of this study was to investigate the effect of AMF and a strain of *T. harzianum* on the growth of tomato and eggplant and against Fusarium wilt of both Solanaceae caused by the fungus *Fusarium oxysporum* f.sp. *albedinis*.

MATERIAL AND METHODS

1- Preparation of plant material

Tomato seeds of the variety Rio grande, and eggplant variety Black Beauty were disinfected with sodium hypochlorite at 5%, rinsed and dried and then left to pre-germinate in alveoli containing peat. Seedlings are grown in plastic pots filled with a mixture of peat and sterile sand. Sterilization is performed in an oven at 250°C for 2 hours to remove the soil microflora. All pots were placed in a greenhouse and watered regularly with tap water.

2- *Trichoderma harzianum* Inoculum

The Tcomp *T. harzianum* strain, isolated from compost (Mouria *et al.*, 2013a) was grown on a potato sucrose agar medium (PSA: 200 g potatoes, 20 g sucrose; Agar-agar 15 g, 1000 ml distilled water) and incubated at 25°C for five days in the dark and five days under continuous light to promote sporulation. The culture surface is then washed with sterile distilled water and the concentration of conidia suspension was adjusted to 10^7 conidia.mL⁻¹.

3- *Fusarium oxysporum* Inoculum

The isolate of *F. oxysporum* f.sp. *albedinis* FA5 was isolated from the date palm tree roots attacked by bayoud (Sghir *et al.*, 2015). The FA5 isolate is cultured on PSA medium and incubated at 28°C in the dark for 7 days. The conidial suspensions were prepared by washing the cultures with sterile distilled water. The concentration is adjusted to 10^5 conidia/ml using distilled water.

4. Inoculation procedure

4-1- mycorrhizal inoculation

The roots of barley plants inoculated with mycorrhizae were rinsed 3 times with distilled water and cut into fragments of 1 to 2 mm in length. Three grams of these fragments have been applied against the root of each tomato and eggplant plant. Control plants were not inoculated with fragments of barley roots.

4-2- *T. harzianum* inoculation

Inoculation is made by soaking roots of tomato and eggplant seedlings coated with their germination substrate in the suspension of *T. harzianum* strain for 30 min (Mouria *et al.*, 2007). The roots of control plants were soaked in sterile distilled water.

4-3- *F. oxysporum* inoculation

The roots of the tomato seedlings and eggplant having reached the stage of two true leaves are stripped from the peat before being inoculated with the conidial suspension of FA5 isolate for 30 minutes, then they are cultivated in the Mamora forest soil.

5- Soil physico-chemical analyzes

The main physico-chemical characteristics of the Mamora forest soil were determined by standard laboratory of soil analyzes ORMVAG (Kenitra, Morocco).

6. Experimental set

The experimental protocol is carried out randomly, eight batches of tomato plants and eight batches of eggplant were performed with six plants for each lot. The pots were then placed in a greenhouse for 3 months at a temperature of 18 to 25°C.

Lot 1: Control plants (T) have not undergone any treatment.

Lot 2: Plants inoculated only with the strain Tcomp (Tc).

Lot 3: Plants inoculated only with AMF (Myc).

Lot 4: Plants inoculated simultaneously with the AMF and Tcomp (Myc + Tc).

Lot 5: Plants inoculated only with FA5 (Foa).

Lot 6: Plants inoculated simultaneously with FA5 and Tcomp (Foa + Tc).

Lot 7: Plants inoculated simultaneously with FA5 and the AMF (Foa + Myc).

Lot 8: Plants inoculated simultaneously with FA5, AMF and Tcomp (Foa + AMF + Tc).

Watering is done every day with distilled water to the plants inoculated with the AMF to promote conditions for the installation of mycorrhiza, and tap water for the other treatments.

7. Evaluation of mycorrhizal parameters

7-1- Root mycorrhization

After three months of inoculation with mycorrhizal fungi, determination of AMF root colonization of tomato plants and eggplant was conducted using the staining technique roots of Phillips and Hayman (1970).

7-2- Spore's extraction

The AMF spores were extracted according to the wet sieving method described by Gerdemann Nicolson (1963).

8- Evaluation of agronomic parameters

8-1- The growth parameters

After 3 months of culture, the roots of tomato plants and eggplant were washed with tap water. At the end of the test the growth parameters, plant height, number of leaves, shoot and root biomass and stem diameter were measured.

8-2- The calculation of dwarfing indices and leaf alteration indices

The leaves alteration degree is noted according to the scale of Douira and Lahlou (1989) below: 0: leaves with healthy appearance; 1: wilting or chlorosis of the cotyledons; 2: loss of the cotyledons; 3: wilting or chlorosis of the true leaves; 4: necrosis of the true leaves; 5: loss of the true leaves. The total score of all the leaves constituted the foliar index. An average was calculated for each lot of plants.

$$L.A.I. = [\sum(i \times x_i)] / (6 \times NtL)$$

LAI: Leaf alteration indice.

i: Leaves appearance notes 0-4.

x_i : Number of leaves with the note i.

NtF: Total number of leaves.

The stem size of all plants is measured at the end of the tests, and the dwarfing indices (D.I.), corresponding to the size reduction of inoculated plants compared to control ones, is determined by the following relation:

$$D.I. = (M-X) / M$$

X: Stem height of the inoculated plants.

M: Average size of the control plants for each substrate.

9- Reisolation of *T. harzianum*

Fragments of thin roots and stem of eggplants and tomatoes inoculated with the strain Tcomp of *T. harzianum* were cut and disinfected with alcohol at 95° for 2 minutes. They were then rinsed several times with sterile distilled water, dried quickly on sterile filter paper, subcultured on the PSA medium and incubated in the dark at 25°C.

10. Reisolation of *F. oxysporum*

Different parts of eggplant and tomato plants inoculated with the strain of *F. oxysporum* FA5 of all treatments were cut and disinfected with alcohol at 95° for 2 minutes. They were then rinsed several times with sterile distilled water, dried quickly on sterile filter paper, subcultured on the PSA medium and incubated in the dark at 25°C.

After a week of incubation, the percentage of colonization of different vegetable parts with *T. harzianum* or *F. oxysporum* is calculated using the following equation.

$$\% Ci = \frac{Ni}{Nt} \times 100$$

% Ci: colonization percentage.

Ni: Number of plants having hosted the pathogen in section i.

Nt: Number of the used plants.

11. Experimental set and Statistical Analysis

The pots were arranged in randomized blocks in a plastic greenhouse. The test was conducted during the months of February-May. Data were analyzed by one-way analysis of variance (ANOVA) and LSD test at 5% level..

RESULTS

1- Study of agronomic parameters

The tomato plants inoculated simultaneously with mycorrhizae and *T. harzianum* showed the best agronomic parameters: the number of flowers: 8; fruit's number: 1.5; leave's number: 14.5; length of the aerial part: 46 cm; length of the root part: 18cm; the fresh weight of the aerial part: 38.5 g and fresh weight of root part 15.1 g (Table 1). Inoculation of plants with the strain FA5 of *F. oxysporum* f. sp. *albedinis* isolated from date palm roots attacked by bayoud showed characteristic symptoms of this disease as wilting and blanching of the leaves (Figure 1). The Figure 2 illustrates the effect of different treatments with *T. harzianum* and mycorrhizae on growth and plant health of tomato plants in the presence and the absence of the pathogen.

Eggplant seedlings inoculated only with the pathogen showed the lowest agronomic parameters (Table 2), including the number of flowers (2.3), leaf's number (6.1), fruit's number (0), length of the aerial part (10.3 cm), root part length (3.4 cm) and the stem diameter (0.5 cm).

Contrary to tomato plants, we didn't observe the characteristic symptoms of the disease caused by *Fusarium oxysporum* f. sp. *albedinis* in eggplant seedlings inoculated with the isolate FA5 of the pathogen (Figure 3).

Co-inoculation of plants with the pathogen, Tcomp and mycorrhizae significantly improved the agronomic parameters including the number of flowers (6.9), fruit number (0.6) and the length of the root part (7.2 cm) (table 2).

Tomato plants inoculated only with *F. oxysporum* f. sp. *albedinis* showed the highest dwarfing indices and leaf alteration indices (DI: 80%, LAI: 0.812) (Table 3). The inoculation of the plants with mycorrhizae or with *T. harzianum* reduced dwarfing indices and leaf alteration indices compared to the control, respectively 2.23% and 0.06 for mycorrhizae and 3.87% and 0.065 for *T. harzianum*. Dual inoculation of tomato plants with *T. harzianum* and mycorrhizae declined both indices (DI: 3.75%, LAI: 0.0210).

Similarly, the inoculation of eggplants with only *F. oxysporum* f. sp. *albedinis* induced the highest DI and LAI, respectively 56% and 0.689 then the co-inoculation of plants with mycorrhizae and Tcomp showed the greatest reduction of these two indices, IR and IAF, respectively 3.45% and 0.0816 for (Table 3).

II- Re-isolation of *T. harzianum* and Foa from tomato plants and eggplant

The re-isolation percentages of Tcomp and FA5 from different parts of the tomato plant and eggplant vary depending on the different treatments (Table 4; Figure 4). The presence of mycorrhizae did not stop the development and installation of *T. harzianum* in the roots of both Solanaceae. The inoculated plants by the pathogen showed continued colonization throughout the plant. The application of the biocontrol agents has limited this extension.

3- Study of mycorrhizal parameters

The microscopic observation of tomato plants and eggplant root fragments treated with mycorrhizae showed the presence of endomycorrhizal structures: hyphae, arbuscules, vesicles and endophytic (Figure 5).

3.1- Mycorrhizal parameters

The results show that eggplant and tomato plants inoculated only with mycorrhizae showed the best mycorrhizal parameters. The presence of *F. oxysporum* f. sp; *albedinis* and *T. harzianum* did not prevent the endomycorrhizal formation in the two Solanaceae. It should be noted that plants non-inoculated with mycorrhizae, in all treatments have noted an absence of endomycorrhizal structures (Table 5).

3.2. Identification of mycorrhizal species

Nine endomycorrhizal species were isolated from the rhizosphere of tomato plants and eight from eggplant. Species identification was made (Figure 6) basing on their morphological characteristics and by reference to the database invam (Barrett and Morton, 2016). These species belongs to genera *Glomus*, *Acaulospora*, *Gigaspora*, *Scutellospora* and *Entrophospora* (Figure 6). The *Glomus* was the most represented with 6 species. Table 6 reports the isolation frequencies of these mycorrhizal species isolated from the rhizosphere of tomato plants and eggplant. The species *Glomus intraradice* was found in the rhizosphere of the two Solanaceae and the isolation frequency of mycorrhizal species varied between 1% and 40%.

Table 1: Effects of *T. harzianum* and mycorrhizae on growth of inoculated and non inoculaed tomato plants with *F. oxysporum* f. sp. *albedinis*.

	Flower number	Fruit Number	Leaves number	A.P.L. (cm)	R.P.L. (cm)	A.F.W. (g)	R.F.W. (g)	S.D (cm)	Fruit Weight (g)	cotyledonary leaves number
T	2.0c	0.9b	3.5c	15c	10c	16.5c	9.1c	0.3c	8c	0b
Tc	6.3b	1.5a	6.7b	25b	16b	35.2b	14.5b	2.3a	11.1b	0b
My	4.5b	1.5a	9.5b	20b	15b	34.1b	17.5a	2.1a	13.2a	0b
Tc+My	8.0a	1.5a	14.5a	46a	18a	38.5a	15.1a	1.7b	14.5a	0.5a
F	1.3d	0.9b	3.5c	13c	12c	10c	10c	1.5b	3c	0b
F+My	2.4c	0.9b	3.1c	12.5c	10.5c	16.5c	12.5c	0.6c	5.6c	0b
F+Tc	3.2b	0.9b	4.5c	14.5c	11.5c	18.3c	14.5b	0.5c	7.5c	0b
F+Tc+My	3.4b	0.8b	5.1c	13.5c	12c	15.4c	15.1b	0.9c	8.1c	0b

A.P.L.: Aerial part lenght. R.P.L.: Root Part lenght. A.F.W.: Aerial fresh weight. R.F.W.: Root fresh weight. S.D : Stem diameter.

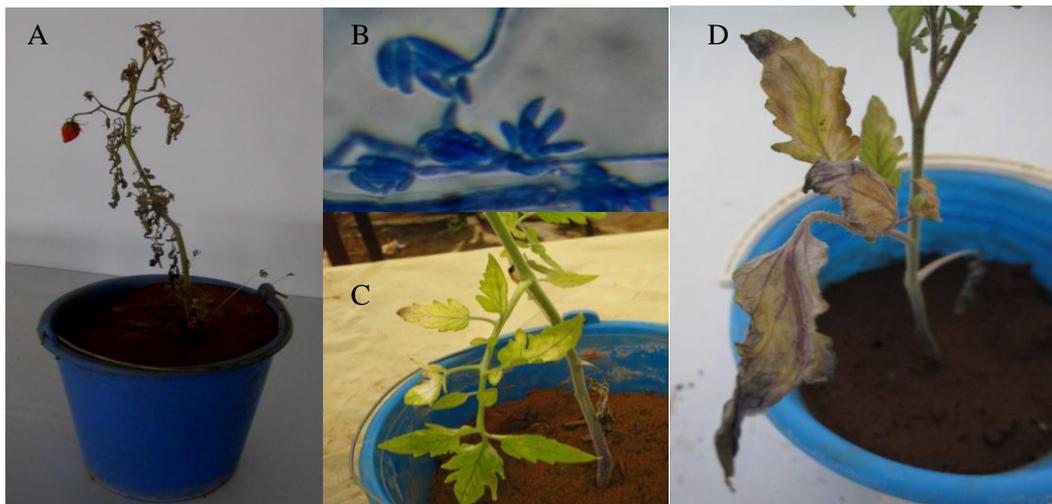


Figure 1. Effect of *F. oxysporum* f.sp. *albedinis* on tomato plants. (A) tomato plant inoculated with *F.o.a*, (B) Spores of *F.o.a*, (C and D) symptoms of *F.o.a* on tomato leaves.



Figure 2. Effects of *T. harzianum* and mycorrhiza on the growth of aerial part (A) and root part of inoculated and non inoculated tomato plants (B) with *F. oxysporum albedinis*.

Table 2. Effects of *T. harzianum* and mycorrhizae on growth of inoculated and non inoculated eggplants plants with *F. oxysporum* f.sp. *albedinis*.

	Flower number	Fruit Number	Leaves number	A.P.L. (cm)	R.P.L. (cm)	A.F.W. (g)	R.F.W. (g)	S.D (cm)	Fruit Weight (g)	cotyledonary leaves number
T	3c	0.3b	8.2c	12.5c	9.4c	16.3d	12.9b	1.4b	15b	0b
Tc	6.4b	0.4b	20.1b	20.4b	16.01b	36.5b	14.3b	1.6b	20.1b	0b
My	7.5b	0.4b	24.3b	42.5a	15.3b	37.1b	19.4a	1.9b	26.5b	0b
Tc+My	10.3a	0.5a	26.5a	40.1a	17.3a	42.1a	18.3a	2.3a	30.4a	0.3 a
F	2.3c	0c	6.1c	10.3c	3.4c	26.4c	8.4c	0.5c	12.4b	0b
F+My	4.5c	0.2b	7.4c	11.4c	5.3c	28.2c	6.3c	0.7c	13.2b	0b
F+Tc	6.1b	0.3b	8.1c	12.5c	6.1c	30.1c	7.1c	0.9c	11.5c	0b
F+Tc+M	6.9b	0.6a	9.1c	13.6c	7.2b	32.4c	9.4c	0.6c	12.5c	0b

A.P.L.: Aerial part length. R.P.L.: Root Part length. A.F.W.: Aerial fresh weight. R.F.W.: Root fresh weight. S.D: Stem diameter.

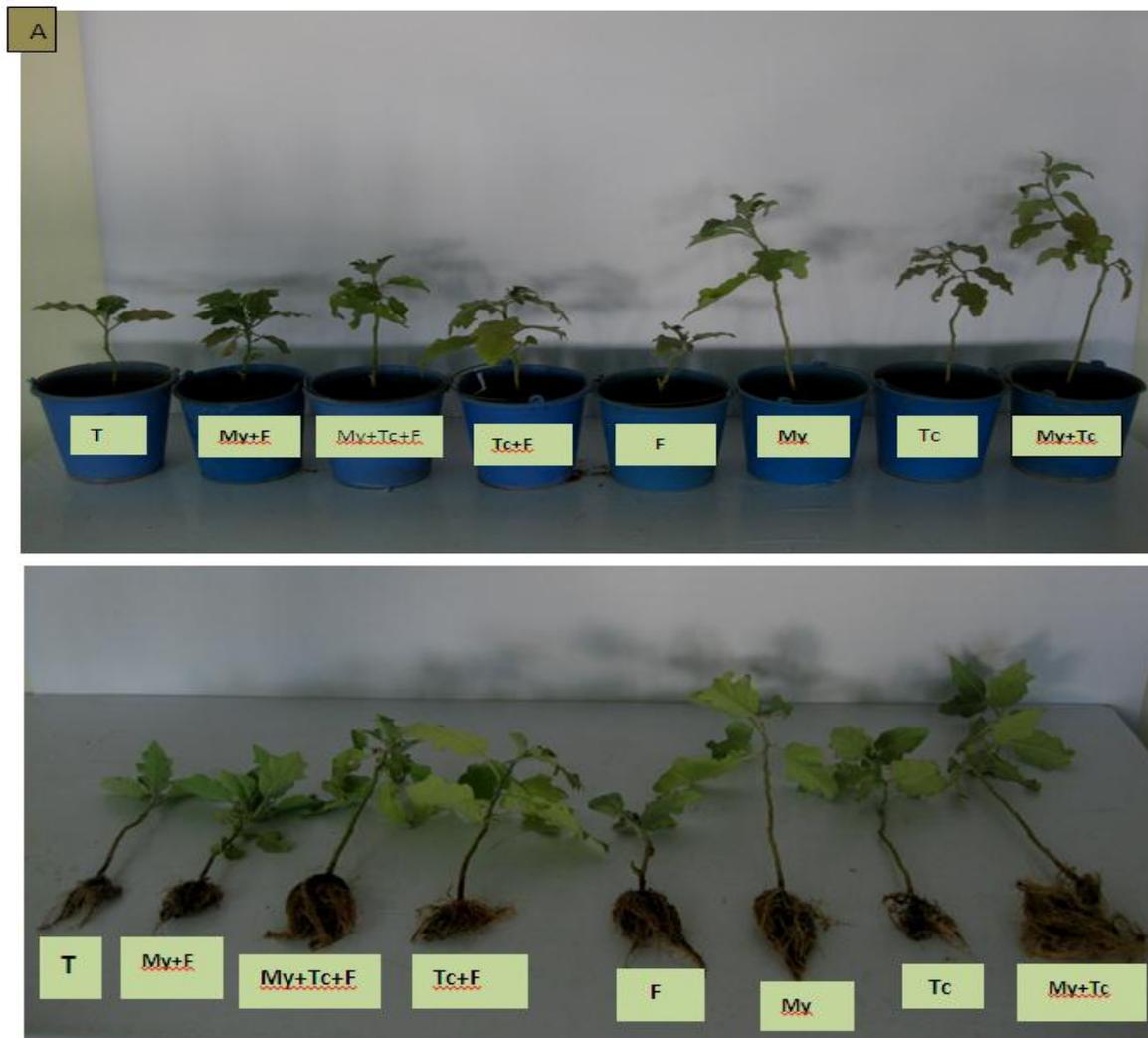


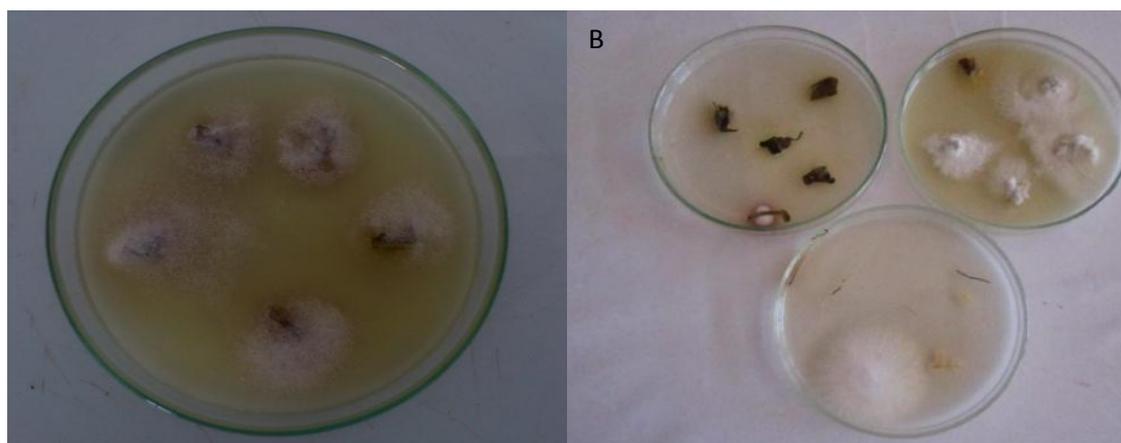
Figure 3. Effects of *T. harzianum* and mycorrhizae on growth (A) and root development (B) of inoculated and non inoculated eggplants with *F. oxysporum* f. sp. *albedinis*.

Table 3. Dwarfing Indices and Leaf alteration indices of eggplant and tomato plants in different treatments.

Treatments	Dwarfing Indices (%)		Leaf alteration indices	
	eggplant	Tomato	eggplant	Tomato
T	6.3c	7c	0.190b	0.160b
Tc	4.01c	3.87c	0.068d	0.065c
Myc	2.79c	2.23c	0.070d	0.060c
Tc+Myc	3.45c	3.75c	0.0816c	0.0210d
F	56a	80a	0.689a	0.812a
F+Myc	19.5b	13.55b	0.387b	0.120b
F+Tc	13.9b	16.34b	0.598b	0.118b
F+Tc+Myc	3.1c	4.5c	0.067d	0.100b

Table 4. Re-isolation of *T. harzianum* and mycorrhizae from different parts of the tomato plant and eggplant. R: Roots; S: Stem; L: Leaves.

		Treatments								
			T	Tc	My	My+Tc	F	F+My	F+Tc	Tc+My+
Tomato	FA5	R	0a	0b	0a	0b	100a	10a	13b	3b
		S	0a	0b	0a	0b	100a	0b	0c	0b
		L	0a	0b	0a	0b	70b	0b	0c	0b
	Tcomp	R	0a	100a	0a	80a	0c	0b	100a	73a
		S	0a	0b	0a	0b	0c	0b	0c	0b
eggplant	FA5	R	0a	0b	0a	0b	100a	12a	15b	6b
		S	0a	0b	0a	0b	95a	0b	0c	0b
		L	0a	0b	0a	0b	76b	0b	0c	0b
	Tcomp	R	0a	100a	0a	100a	0c	0b	100a	80a
		S	0a	0b	0a	0b	0c	0b	0c	0b



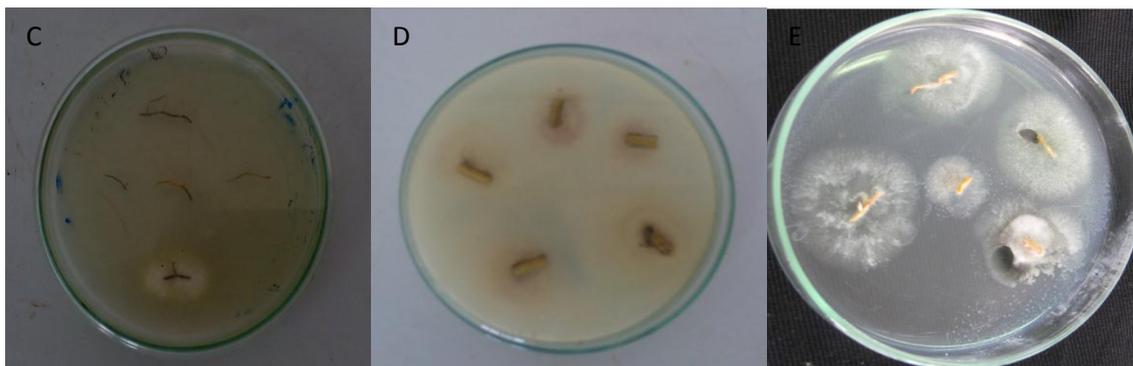


Figure 4. Isolation of Tcomp and FA5 from different parts of the tomato plant and eggplant. (A) reisolation of FA5 from tomato leaves, (B) reisolation of Tcomp from tomato roots, (C) reisolation of FA5 from eggplant roots, (D) reisolation of FA5 from the tomato stem, (E) reisolation of Tcomp from eggplant roots.

Table 5: mycorrhizal parameters of tomato and eggplant plants according to the different treatments.

		Mycorrhizal parameters				
		Mycorrhizal frequency %	Mycorrhizal intensity%	Arbuscular content %	Vesicular content %	Spores density
Tomato	My	85a	78a	95a	56a	300a
	My+F	35b	63b	80a	39b	260b
	My+Tc	40b	56b	75b	47b	220b
	My+Tc+F	20c	48b	41b	46b	180c
	T	0d	0c	0c	0c	0d
	F	0d	0c	0c	0c	0c
	Tc	0d	0c	0c	0c	0c
Eggplant	My	91a	80a	75a	60 a	340a
	My+F	51b	45b	72a	58a	280b
	My+Tc	47b	38b	60b	51a	246b
	My+Tc+F	28c	36b	62b	41b	190c
	T	0d	0c	0c	0c	0d
	F	0d	0c	0c	0c	0d
	Tc	0d	0c	0c	0c	0d

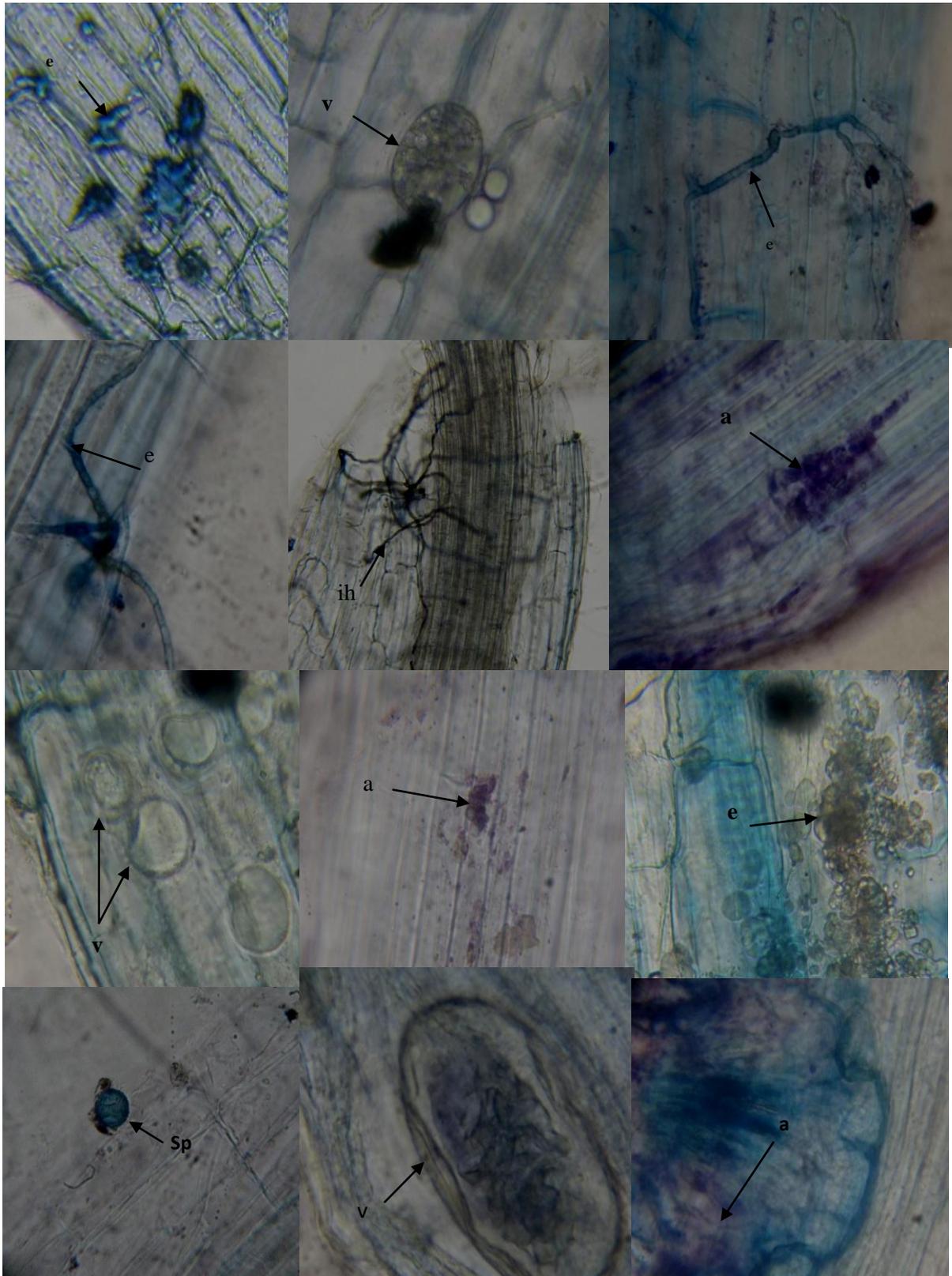


Figure 5: Different endomycorrhizal structures in eggplants and tomatoes plants inoculated with mycorrhizae. Sp: spore, e: endophyte, ih: internal hyphae, a: arbuscule, v: vesicle.

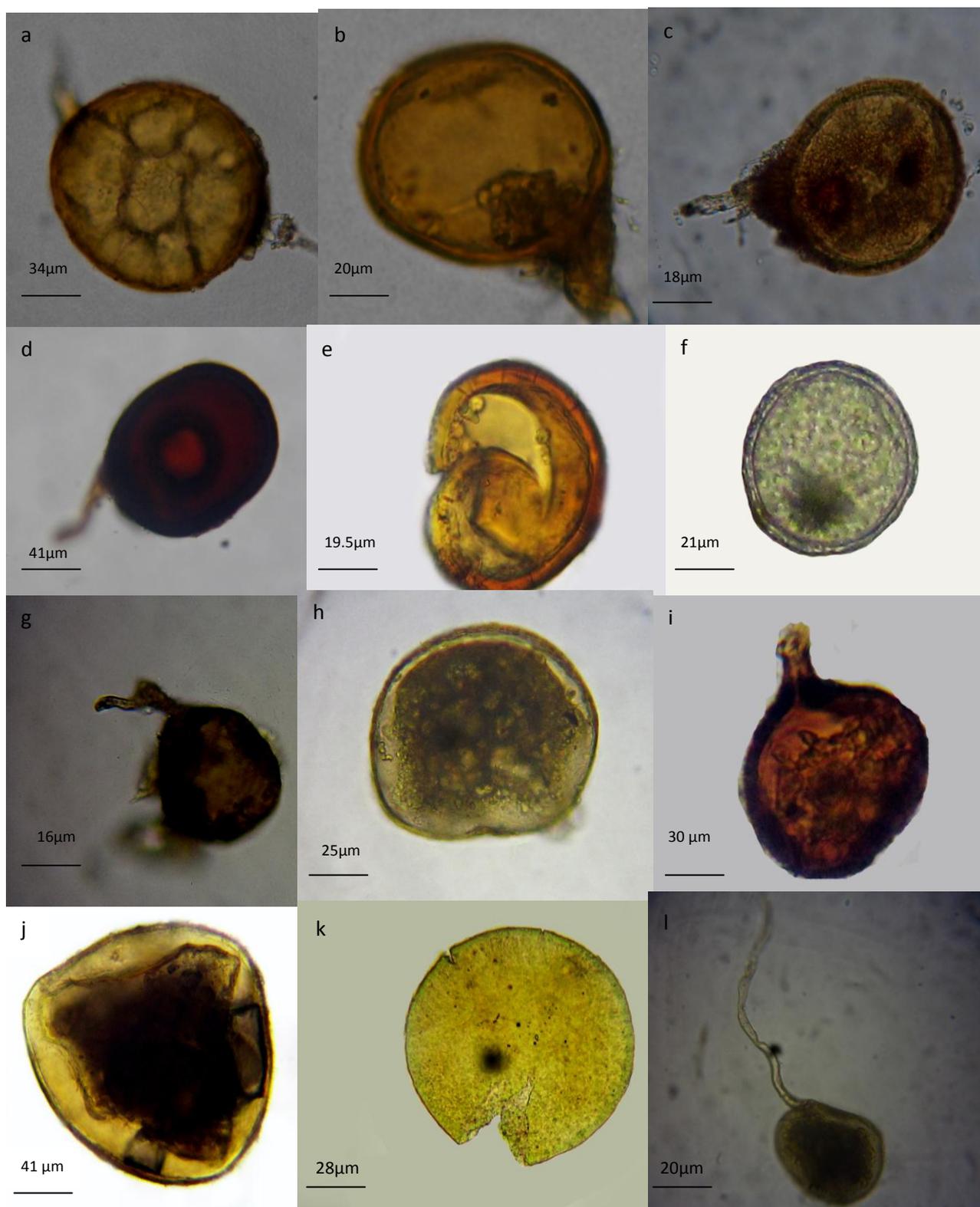


Figure 6. Endomycorrhizal species isolated from the rhizosphere of eggplant and tomato plants treated with mycorrhizae. a: *Glomus* sp1, b: *Acaulospora* sp1, c: *Gigaspora* sp1, d: *Scutellospora* sp1, e: *Glomus* sp2, f: *Scutellospora* sp1, g: *Gigaspora* sp2, h: *Glomus badium*, I: *Glomus* sp1, j: *Acaulospora* sp1, k: *Glomus* sp2, L: *Entrophospora* sp1.

Table 6. Isolation frequency (%) endomycorrhizal species from the rhizosphere of tomato and eggplant seedlings.

Species	Tomato	Eggplant
<i>Glomus</i> sp1	40a	
<i>Glomus</i> sp2	–	30a
<i>Acaulospora</i> sp1	5b	–
<i>Acaulospora</i> sp1	–	8c
<i>Glomus</i> sp3	10b	–
<i>Glomus</i> sp4	–	15b
<i>Gigaspora</i> sp1	9b	–
<i>Gigaspora</i> sp2	–	12b
<i>Glomus badium</i>	7b	–
<i>Glomus intraradice</i>	2b	12b
<i>Gigaspora</i> sp1	6b	–
<i>Scutellospora</i> sp1	–	1c
<i>Scutellospora</i> sp2	3b	–
<i>Entrophospora</i> sp3	1b	–
<i>Acaulospora foveata</i>	–	2c
<i>Acaulospora</i>	–	3c

DISCUSSION

The mycorrhizal inoculum is increasingly applied in the production and cultivation of vegetable (Janas *et al.*, 2002) due to its positive effects on yield parameters. Edathil *et al.* (1994) observed the natural colonization of tomato and eggplant roots by the AMF in a pot culture. Arbuscular mycorrhizae are associated with 83% of broadleaf and 79% of monocotyledonous (Trappe, 1987). All gymnosperms are reported as mycorrhizal (Newman and Reddell, 1987). Mycorrhizae play an important role in ion absorption from the soil, especially phosphorus, zinc and copper (Ortas *et al.*, 2002).

In this study, we showed the effect of mycorrhizae against *F. oxysporum* FA5 on eggplant and tomato plants. Indeed, the application of mycorrhizae only showed a strong inhibition of dwarfing indices and leaf alteration indices in both species of Solanaceae.

In this sense, Ozgonen *et al.* (2007) showed that the application of endomycorrhizal fungi such as *Glomus mosseae*, *G. etunicatum*, *G. fasciculatum* and *Gigaspora margarita* strengthened the development of pepper plants and reduces the incidence of mildew disease

on this plant. This same beneficial effect was made during the application of mycorrhizal species, *Glomus mosseae*, against *Verticillium* wilt of eggplant and tomato (Karagiannidis *et al.*, 2002). These authors also reported that this species has also improved nutrient uptake and growth in these two plants.

Indeed, our results showed that the application of the AMF has caused stimulation of agronomic parameters of tomato plants and eggplant. Bhuiyan (2013) said that the significant absorption of nutrients by the plants after application of mycorrhizae increase yield and plants biomass. Inoculation of pepper plants with *Glomus deserticola* increases phosphorus absorption and reduces the deleterious effect of *Verticillium dahliae* on the yield of this plant (Garmendia *et al.*, 2004). This reduction in the pathogen effect is also observed when applying *Glomus mosseae* on tomato plants against *Phytophthora nicotianae var. parasitica* C. (Cordier *et al.*, 1996).

The first contact between the AMF and plant roots induced mechanisms of defense by increasing phenolic compounds (Morandi, 1996, Mukerji, 1999). The increase in the phenolic content of the cell wall of mycorrhizal roots due to AMF secretions strengthens the cell wall and prevents the invasion of pathogens (Graham, 1991). Another study showed that the combination of organic matter, humic acid and whey, with mycorrhizal fungi (AMF), improves the growth parameters of tomato, pepper and eggplant and increases their resistance to diseases (Demir *et al.*, 2015). Similarly, the combination of a rhizobacteria and AMF has improved the use of fertilizers and increase the efficiency of their nutrients (Adesemoye *et al.*, 2009). The study of Khalil *et al.* (2012) showed that tomato plants treated with the AMF are able to overcome the attacks of *Meloidogyne* compared to untreated plants (Azcón-Aguilar and Barea, 1997).

Furthermore, we have shown in this work that inoculation of tomato plants and eggplant with *T. harzianum* isolated from compost, strongly decreased the effect of *F. oxysporum* on both Solanaceae judging by the decrease of the dwarfing indices and leaf alteration indices. Indeed, this biological control agent is very effective against seedling burns diseases of eggplant and also promotes seeds germination (Meah *et al.*, 2004).

Jee and Kim (1987) reported that *T. harzianum* is the best antagonist against soil pathogens *in vitro*. Indeed, it is an effective control agent against *Phytophthora* that causes the damping off of cucumber (Tehroni and Nazari, 2004). Montaser (2014) showed that *T. viride* and *T.*

harzianum are the best biological control agents for their ability to reduce significantly the impact of diseases and increase plant yield.

Indeed, the application of *Trichoderma* as a biological control agent is based on several direct and indirect mechanisms that can work alone or in synergy, such as the production of volatile and non-volatile secondary metabolites that inhibit pathogens (Harman and Björkman, 1998; Monte, 2001; Howell, 2003; Benitez *et al.*, 2004, Harman, 2006). This process of inhibition is called antibiosis (Hjeljord and Tronsmo, 1998).

But the main mechanism of antagonism of *Trichoderma* species is mycoparasitism. This is the lytic activity which is responsible for the expression of mycoparasitism against several fungal pathogens (Chet, 1987). Wu *et al.* (2008) described the potential hyperparasite of *T. harzianum* on *S. sclerotiorum*, as a redoubtable eggplants pathogen which is manifested by inhibition of mycelial growth and sclerotia production.

Many researchers have reported the antagonistic activity of *Trichoderma* isolates against the plant pathogens, particularly against fungal pathogens such as telluric *Rhizoctonia solani*, *Fusarium oxysporum*, *Sclerotium rolfsii* and *Verticillium dahliae* (Lo *et al.*, 1996; Tran 1998; Bari *et al.*, 2000; Shamsuzzaman *et al.*, 2003; Ngo *et al.*, 2006; Shalini *et al.*, 2006; Mouria *et al.*, 2013b). In our work, the eggplant and tomato plant inoculated with *T. harzianum* also showed higher agronomic parameters compared to control plants. Indeed, *Trichoderma* strains often enhance the growth and yield of plants and the development of their roots (Harman *et al.*, 2004; Altintas and Bal, 2005; Mouria *et al.*, 2007; Bal & Altintas, 2006). Indeed, the work of Srivastava (2004) and Mouria *et al.* (2007) demonstrated that the colonization of plant roots by *Trichoderma* strains and their competitiveness in the rhizospheric soil are responsible for improving the growth and development of the roots of these plants.

The improvement of seed germination after inoculation with species of *Trichoderma* was also reported in many species (Kumar and Dubey, 2001). Other studies have indicated that the reduction of the incidence of the disease and seed germination increase leads to a higher yield when treating seeds and soil with *Trichoderma* species (Dubey and Patel, 2001 ; Poddar *et al.*, 2004).

Our study allowed us to note that the combined application of mycorrhizae and *T. harzianum* improved agronomic parameters of tomato plants and eggplant and has greatly reduced leaf alteration indices and dwarfing at very low levels compared to other treatments. In this sense, the co-inoculation has given weaker DI and LAI. Sghir *et al.* (2014) also highlighted the effect of double inoculation with *T. harzianum* and AMF on improving the growth of date palm seedlings and also on Fusarium wilt of these plants caused by the strain of *F. oxysporum* used in this work. Nzanza (2011) showed that the application of *T. harzianum* and AMF delays the incidence of verticillium wilt in probably mid-season due to the capacity of these fungal species against soil pathogens. Similarly, the application of *T. harzianum* combined with the AMF showed better protection of cucumber plants against damping off disease caused by *Rhizoctonia* (Chandanie *et al.*, 2009).

The effect of AMF and *T. harzianum* against Fusarium wilt of tomato plants and eggplant could be explained by acquired resistance which is probably due to the pre-activation of phenolic compounds, especially phytoalexins, flavonoids and associated isoflavonoids, secreted by the AMF in roots (Karagiannidis *et al.*, 2002), which blocks the entry points at root level and consequently suppresses the development of the pathogen in the rhizosphere by antibiosis (Tahmatsidou *et al.*, 2006). The beneficial effects of vesicular arbuscular mycorrhizae (VAM) and *T. viride* as a means against pathogenic fungi *R. solani* and *F. solani* that cause infections and decrease the growth and quality of sugar beet have been demonstrated by Ali and Hussein (2009). The use of mycorrhizal species *G.moseae* and *T. harzianum* reduced the severity of Verticillium wilt caused by *V. dahliae* (Azcón-Aguilar and Barea, 1996; Inbar *et al.*, 1996; Akköprü *et al.*, 2005; Arıcı 2009).

Finally, our results showed that inoculation of tomato plants and eggplant with *T. harzianum* reduced the colonization of their roots by AMF without deleting it, but the two biocontrol agents exist at the level of roots. Green *et al.* (1999) reported the same effect, colonization of cucumber roots by *Glomus intraradices* was significantly reduced in the presence of *T. harzianum*.

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