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Alleviation of Rhizoctonia root rot damage in common bean by some arbuscular mycorrhizal fungi

Nahid Moarrefzadeh✉, Hadi Khateri, Saeed Abbasi

Department of Plant Protection, College of Agriculture, Razi University, Kermanshah, Iran.✉n.moarrefzadeh@razi.ac.ir

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Abstract

Rhizoctonia root rot of common bean (*Phaseolus vulgaris*) caused by *Rhizoctonia solani* is among the most important soil-borne fungal diseases worldwide. In this study, nine arbuscular mycorrhizal fungi (AMF) including *Acaulospora longula*, *Funneliformis mosseae*, *Gigaspora margarita*, *Glomus caledonium*, *G. claroideum*, *G. etunicatum*, *G. fasciculatum*, *G. versiform* and *Rhizophagus irregularis* were evaluated for their effect on some growth traits and inhibition of *R. solani* in bean plants under greenhouse conditions. Six AMF species (*F. mosseae*, *G. claroideum*, *G. etunicatum*, *G. margarita*, *G. caledonium* and *G. versiform*) significantly reduced the disease severity index and the first four of these also reduced the incidence of disease compared with the infected control. The lowest disease severity and incidence were obtained by *F. mosseae* and *G. claroideum*, respectively. Compared with the infected control, the root length was significantly improved by all AMF. The other growth traits were also significantly improved by all AMF species with some exceptions as follows: root wet and dry weights (except *G. fasciculatum*), shoot wet weight (excep *G. versiform*), shoot length (except *G. claroideum*, *G. versiform* and *G. etunicatum*) and shoot dry weight (except *G. etunicatum*, *G. fasciculatum*, *G. caledonium* and *G. margarita*). *Glomus fasciculatum* had the highest root colonization. According to the results of this study, many AMF fungi improved plant growth and partially compensated for Rhizoctonia root rot on common bean, and they could be considered as good candidates for studying the biological control of this disease under field conditions.

Keywords: *Rhizoctonia solani*, Endomycorrhizae, Disease severity, Incidence, *Glomus*

کاهش خسارت بوته‌میری رایزوکتونیایی در لوبیا توسط برخی قارچ‌های میکوریزی دارسانه‌ای

ناهید معرف‌زاده✉، هادی خاطری، سعید عباسی

گروه گیاه‌پزشکی، دانشکده کشاورزی، دانشگاه رازی، کرمانشاه، ایران. ✉n.moarrefzadeh@razi.ac.ir

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چکیده

بیماری بوته‌میری رایزوکتونیایی لوبیا (*Phaseolus vulgaris*) در اثر *Rhizoctonia solani* جزو مهم‌ترین بیماری‌های قارچی خاکزاد این محصول در اکثر مناطق کشت در سراسر جهان است. در تحقیق حاضر اثر نُه گونه قارچ میکوریزی دارسانه‌ای (AMF) شامل *Acaulospora longula*، *Funneliformis mosseae*، *Gigaspora margarita*، *Glomus caledonium*، *G. claroideum*، *G.etunicatum*، *G. fasciculatum*، *G.versiform* و *Rhizophagus irregularis* روی مهار بوته‌میری رایزوکتونیایی و برخی صفات رشدی لوبیا در حضور بیمارگر در گلخانه بررسی شد. شش گونه AMF (*F. mosseae*، *G. margarita*، *G. claroideum*، *G. etunicatum* ، *G. versiform* و *G. caledonium*) سبب کاهش شاخص بیماری شدند و از میان آنها، چهار گونه اول، وقوع بیماری را نیز نسبت به شاهد بیمار به‌طور معنی‌داری کاهش دادند. کمترین شدت و وقوع بیماری به ترتیب با کاربرد *F. mosseae* و *G. claroideum* به دست آمد. در مقایسه با شاهد بیمار، همه گونه‌های AMF سبب افزایش معنی‌دار طول ریشه و علاوه بر آن وزن تر و وزن خشک ریشه (به‌استثنای *G. fasciculatum*) شدند و وزن تر اندام هوایی (به‌استثنای *G. versiform*)، طول اندام هوایی (به‌استثنای *G. claroideum*، *G. versiform* و *G. etunicatum*) و وزن خشک اندام هوایی (به‌استثنای *G. etunicatum*، *G. fasciculatum*، *G. caledonium* و *G. margarita*) را نیز افزایش دادند. بیشترین میزان کلنیزاسیون میکوریزی ریشه نیز در تیمار *G. fasciculatum* مشاهده شد. طبق نتایج این تحقیق بسیاری از قارچ‌های AMF سبب بهبود رشد گیاه و جبران بخشی از خسارت بوته‌میری رایزوکتونیایی لوبیا گردیدند و این عوامل، گزینه‌های مناسبی برای بررسی کنترل بیولوژیکی این بیماری در شرایط مزرعه هستند.

کلمات کلیدی: *Rhizoctonia solani*، اندومیکوریزها، شدت بیماری، وقوع، *Glomus*

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Introduction

Common bean (*Phaseolus vulgaris* L., from the family Fabaceae) is an important source of protein supply and energy for humans (Bitocchi *et al.* 2012). This plant is invaded by a variety of plant pathogens especially fungi, that are able to reduce the yield by 80% and in some cases up to 100% (Gupta *et al.* 2018). *Rhizoctonia solani* Kuhn (teleomorph: *Thanatephorus cucumeris* (Frank) Donk) is one of the most important soil-borne plant pathogens of common bean that has been reported from most growing areas around the world (D'Aes *et al.* 2011; Erper *et al.* 2011). This pathogen is very aggressive on different bean cultivars and reduces the yield and quality by causing seed rot, damping-off, hypocotyl rot, stem necrosis and root rot (Abawi *et al.* 1985).

The control of *R. solani* in different environmental conditions is difficult due to the high diversity in the populations of this pathogen, its ecological behavior and ability of saprophytic activity, widespread distribution in soils, the wide host range and the high survival rate of sclerotia (Agrios 2005; Groth & Bond 2006; Nawrocka *et al.* 2018). These are of the important reasons that why agricultural management practices including sanitation, crop rotation and soil solarization are not effective enough to control this pathogen (Aljawasim *et al.* 2020). In addition, no acceptable resistant varieties are currently available against this pathogen (Valentín Torres *et al.* 2016). Despite the effects of some chemical fungicides for controlling *R. solani*, they are not recommended due to their adverse side-effects on the non-target organisms, development of resistant populations (Daroodi *et al.* 2021), increasing the production costs, causing serious risks for human health and the environmental pollutions (Madhavi *et al.* 2018). Therefore, the use of an integrated disease management strategy is necessary for alleviating the pressure caused by this destructive pathogen (Schwartz 2011). Biological control, as a safe and environmentally friendly method could be considered as one of the important components of this integrated management strategy against this disease (Aljawasim *et al.* 2020; Daroodi *et al.* 2021; Haque & Parvin 2021; Heflish *et al.* 2021). Beneficial organisms in the rhizosphere form a primary barrier against pathogens invading the roots (Akhtar *et al.* 2010). Therefore, the use of microorganisms that can grow in the rhizosphere as biocontrol agents seems to be an ideal method for controlling soil-borne pathogens (Atwa 2018). Among such microorganisms, arbuscular mycorrhizal fungi (AMF) have received special attention. AMF are a group of beneficial soil microorganisms that have a symbiotic relationship with about 80% of vascular plants and are completely dependent on nutrients provided from the living root systems of plants (Jacott *et al.* 2017). In return for this symbiotic relationship, AMF provide their plant partners with many benefits such as increasing the germination rate and growth of the plants and enhancing the absorption of water and nutrients (Rouphael *et al.* 2015; Jacott *et al.* 2017). Mycorrhizal symbiosis also leads to increased resistance in plants against various abiotic stresses (Yao *et al.* 2002) and a wide range of plant pathogens, including fungal pathogens (Li *et al.* 2021; Moarrefzadeh *et al.* 2021b, 2021a; Wu *et al.* 2021). However, the beneficial effects of AMF are variable and sometimes contradictory results have been obtained in various studies. In some cases, root colonization by AMF has had no effect in reducing the disease (Davis 1980; Bååth & Hayman 1984) and even in some cases the disease severity was increased (Davis *et al.* 1979).

Despite numerous studies on the role of AMF in reducing the incidence and severity of disease in plants infected by *Rhizoctonia* and their beneficial effects on the growth and development of many plants such as potatoes (Mohammed *et al.* 2020), watermelon (Wu *et al.* 2021) and tomato (Aboelmagd 2021), there are few reports available about their effects on *R. solani* infections on common bean (Hafez *et al.* 2013; Matloob & Juber 2013; Nasir Hussein *et al.* 2018). In addition, many AMF species have not yet been studied for their potential effects against plant pathogens such as *R. solani* in common bean. Therefore, the present study was performed to investigate the potential inhibitory effects of nine different AMF species against *Rhizoctonia* root rot in common bean and also compare their potential effects for improving the growth parameters of common bean plants at the presence of the pathogen. Another goal of this study was to find out whether there is a correlation between the rates of root colonization by these species with the severity of *Rhizoctonia* root rot on common bean plants**.**

Materials and methods

Preparing the pathogen inoculum

An isolate of *R. solani* AG-2, which had been previously identified based on morphological characteristics and its pathogenicity had been confirmed on common bean, was obtained from the fungal collection of Plant Protection Department (Faculty of Agriculture, Razi University, Kermanshah, Iran). This isolate was grown in Petri dishes containing potato dextrose agar (PDA) at 25 °C. For propagating the inoculum of this pathogen, 50 grams of millet seeds were washed and then soaked in 150 ml distilled water in 250 ml flasks for 24 hours. After removing the excess water, the seeds were sterilized by autoclave (two times at 24 hours intervals). Five mycelial discs were cut from the growing edge of *R. solani* colony on PDA and transferred to flasks containing millet seeds. The flasks were kept at 25 °C for 2 weeks, and when the seeds were completely colonized by the fungal hyphae, they were used as pathogen inoculum in greenhouse tests (Ardalan *et al.* 2017).

Preparing the mycorrhizal fungi

Nine species of mycorrhizal fungi including *Acaulospora longula, Funneliformis mosseae (syn. Glomus mosseae), Gigaspora margarita, Glomus caledonium, G. claroideum, G.etunicatum, G. fasciculatum, G. versiform and Rhizophagus irregularis* (Synonyms: *Rhizophagus intraradices* and *G. intraradices*) were obtained as commercial formulations from Turan Biotechnology Company (Shahroud, Iran).

Greenhouse experiments

The potential of AMF species in controlling *R. solani* pathogen and their possible effects on the growth parameters of common bean at the presence of the pathogen were evaluated in a pot experiment in greenhouse. This test was based on a completely randomized design with eleven treatments and six replications (Table 1). Common bean seeds (variety Early Khameneh) were surface disinfected by immersing them in 70% ethanol and then 0.5 % sodium hypochlorite (one minute for each), and then thoroughly washed by sterile distilled water for five times. The seeds were imbibed for 3 h in sterile water and germinated for two days by incubating them on top of a clean flat plastic strainer inside a closed humid container at room temperature. The seeds were not in direct contact with the layer of water at the bottom and washed by spraying sterile water thrice a day.

Autoclave-sterilized peat moss and perlite (at volumetric ratio of 1:2) was used as potting substrate. For each AMF species, 36 grams of the commercial inoculum (containing 100 fungal spores per gram) was considered for each 450 ml plastic pot (eight cm diameter and 13 cm height). The AMF inocula were mixed well with the potting substrate before adding them to the pots at the time of sowing the seeds. In order to facilitate the subsequent addition of the pathogen, a plastic tube (15 cm in length and 1 cm diameter) was placed in the center of each pot (Jasem *et al.* 2018), and the prepared mixtures (AMF inocula in potting substrate) were added to about three cm below the top of the pots. Four germinated bean seeds were placed in each pot and then covered with a two cm layer of the same mixtures. After germination, three seedlings were kept in each pot. In both healthy and infected control treatments, the potting substrate was used without adding mycorrhizal inocula (Moarrefzadeh *et al.* 2021b).

Two weeks after sowing the seeds, the plastic tubes that had been previously placed in the center of pots were removed and 50 milligrams of millet seeds colonized by *R. solani* and thoroughly mixed with 10 g sterile vermiculite, was added into the empty space in each pot. In healthy control treatment, only sterile vermiculite and uninfested millet seeds were added.

Inoculated plants were placed randomly in greenhouse conditions at 25 ± 5 °C and irrigated daily with a very gentle stream of water containing 100 ppm of a complete fertilizer (NPK + TE, 18-18-18, Fermolife, Baharan Co., Isfahan, Iran).

Evaluation of bean root rot disease

Two weeks after inoculating by the pathogen (one month after sowing bean seeds), when clear symptoms of disease were seen in the infected control treatment, the disease incidence and severity were evaluated in different treatments according to the method described by Yildirim & Erper (2017). This method uses a 0-4 scale as follow:

0) Seedlings are healthy without disease symptoms; 1) very little superficial lesions on the roots or on the hypocotyls; 2) deep and large lesions on the roots or on the hypocotyl; 3) severe root rot, lesions surrounding hypocotyl, partially restricted root length and 4) complete root rot and seedling death occur. The disease severity index (DSI) was calculated using the following formula (Silva *et al.* 2020): DSI (%) = [Σ (degree of the scale × frequency) / (total number of units evaluated × maximum degree of the scale)] × 100.

Disease incidence (DI) was determined as follows (Ali & Nadarajah 2013): DI (%) = (Number of infected plants / total number of observed plants) × 100.

Growth parameters of mycorrhizal plants at the presence of the pathogen

After disease evaluation, the plants of different treatments were carefully uprooted from the pots. The roots were washed and the growth parameters of bean plants including the fresh and dry weights and length of both the roots and shoots were measured. The data were analysed by the SAS software (version 9.4).

Duncan's multiple range test was used to compare the means and the statistical probability level was considered as 5% in all analyses.

Root colonization by mycorrhizal fungi

The colonization of common bean roots by AMF was assessed as described by Phillips & Hayman (1970) with a slight change. Briefly, 0.1 gram of young roots was rinsed with distilled water, cut into 1 cm pieces and placed in 5 ml tubes containing 3 ml of 10% KOH. The samples were placed in a hot water bath (90 °C) for one hour. After rinsing thoroughly by water, the roots were immersed in 1% hydrochloric acid for 5 minutes and without further rinsing, they were stained for one hour in 0.01% acid fuchsin in lactoglycerol made of 1:1:1 lactic acid, glycerin, and water. The samples were then placed in a hot water bath (90 °C) for one more hour. The stained roots were kept in lactoglycerol without acid fuchsin for 30 minutes. The stained roots (100 pieces from each replication) were evaluated with light microscopy at magnifications of up to 400X for the presence of AMF-related organs. After counting the mycorrhizal and non-mycorrhizal roots, the percentage of root colonization was determined as described by Sohrabi *et al.* (2015).

Results

The effect of AMF on disease severity index and disease incidence

There were significant differences between the endomycorrhizal treatments for their effects on disease incidence and disease severity index in greenhouse conditions. All tested AMF species excluding *G. fasciculatum*, *A. longula* and *R. irregularis*, significantly reduced the disease severity index compared to the infected control at 5% probability level (Figure 1). The lowest disease severity indices were observed by four AMF species including *F. mosseae*, *G. claroideum*, *G. versiform* and *G. etunicatum* that in comparison with the infected control, reduced the disease severity index by 49.3, 41.3, 38.7 and 34.7%, respectively, and placed in the same statistical group. The next two AMF species showing significantly lower disease severity than the infected control were *G. margarita* and *G. caledonium* that reduced the disease severity by

28% compared with the infected control (Figure 1). Among the endomycorrhizal treatments, four species including *G. claroideum*, *G. etunicatum*, *F. mosseae* and *G. margarita,* significantly reduced the disease incidence compared to the infected control by 39.3, 28.6, 21.4 and 21.4%, respectively.

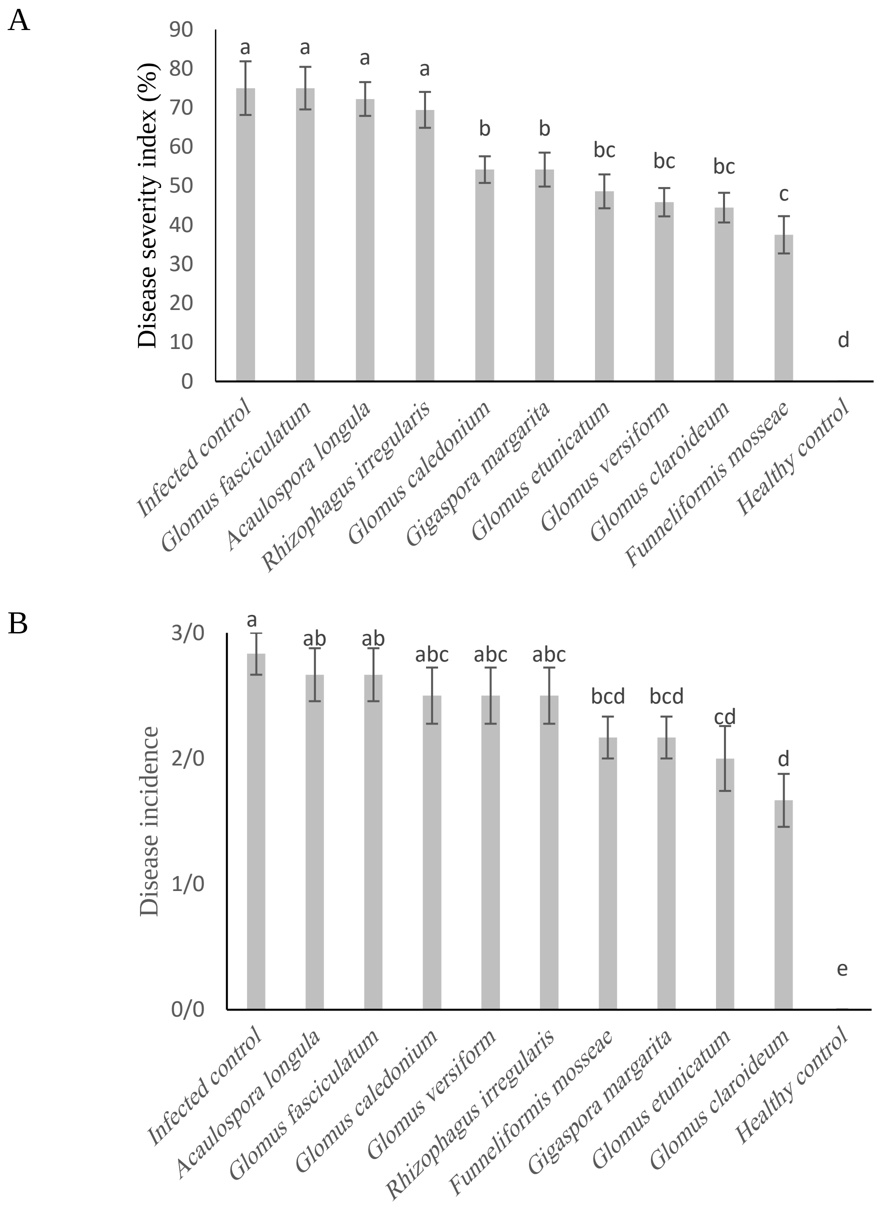

Figure 1. Effect of arbuscular mycorrhizal fungi treatments on disease severity index (A) and disease incidence (B) in common bean (*Phaseolus vulgaris*) plants inoculated by *Rhizoctonia solani* in greenhouse. The means were compared using Duncan's multiple range test at 5% probability level. Bars with common letters are not significantly different.

Effect of AMF on shoot growth parameters at the presence of the pathogen

There were significant differences between the endomycorrhizal treatments for their effects on shoot growth parameters of common bean plants. The fresh weight of shoots was significantly increased by all tested AMF species (except *G. versiform*), and the highest effect was related to *F. mosseae* and *G. claroideum* that increased this index by 33.3% compared to the infected control. All tested AMF species excluding *G. claroideum*, *G. versiform* and *G. etunicatum*, increased the shoot length and the highest effect was related to *G. fasciculatum* and *G. margarita* with 8.7% increase compared to the infected control. The dry weight of shoots was also significantly increased by *F. mosseae*, *G. claroideum*, *G. versiform*, *A. longula* and *R. irregularis*, which were at the same statistical group with the healthy control. Among these species, *G. claroideum* and *F. mosseae* showed the highest shoot dry weights that were respectively 30.4 and 26.47% more than the infected control.

Table 1. Effect of arbuscular mycorrhizal fungi on shoot growth parameters of common bean plants at the presence of the pathogenic fungus *Rhizoctonia solani*.

Treatment	**Stem length (cm)**	**Shoot wet weight (g)**	**Shoot dry weight (g)**
Acaulospora longula	23.7^{cd}	6.2^{e}	1.16^{bcd}
Funneliformis mosseae	23.9^{cd}	8.4^{b}	1.29^{ab}
Gigaspora margarita	24.7^{bc}	6.6^{d}	1.09^{def}
Glomus caledonium	24.5^{bcd}	7.6^{c}	1.14^{cde}
Glomus claroideum	21.8^{f}	8.4^{b}	1.33^{a}
Glomus etunicatum	23.5^{de}	7.6^{c}	0.97^{f}
Glomus fasciculatum	25.2^{b}	7^{d}	0.98^{f}
Glomus versiform	22.6^{ef}	5.4^{f}	1.16^{bcd}
Rhizophagus irregularis	24.4^{bcd}	6.6^{d}	1.24^{abc}
Infected control	22.5^{ef}	5.6^{f}	1.02^{ef}
Healthy control	29.1^{a}	9.3^{a}	1.23^{abcd}

The values are averages of six replications. Means with common letters are not significantly different at $p \leq 0.05$ according to Duncan's multiple range tests.

Table 2. Effect of arbuscular mycorrhizal fungi on root growth parameters of common bean plants at the presence of the pathogenic fungus *Rhizoctonia solani*.

Treatment	**Root length (cm)**	**Root wet weight (g)**	**Root dry weight (g)**
Acaulospora longula	11.6^{g}	0.54^{de}	0.13^{cd}
Funneliformis mosseae	15.2^{ab}	0.73^{a}	0.17^{a}
Gigaspora margarita	12.4^{ef}	0.67^{ab}	0.13^{cd}
Glomus caledonium	15^{ab}	0.59^{cd}	0.12^{e}
Glomus claroideum	13.2^{de}	0.64^{bc}	0.16^{b}
Glomus etunicatum	14.3^{bc}	0.71^{a}	0.13^{d}
Glomus fasciculatum	12^{fg}	0.49^{ef}	0.1^{f}
Glomus versiform	12.8^{def}	0.67^{ab}	0.17^{ab}
Rhizophagus irregularis	13.6^{cd}	0.71^{a}	0.14^{c}
Infected control	10.7^{h}	0.47^{f}	0.1^{f}
Healthy control	15.3^{a}	0.72^{a}	0.14^{c}

The values are averages of six replications. Means with common letters are not significantly different at $p \leq 0.05$ according to Duncan's multiple range tests.

Effect of AMF on bean root growth parameters at the presence of the pathogen

Treatments by all AMF significantly increased the length of roots and the highest effect was related to *F. mosseae* and *G. caledonium* that were 36.4% higher than the infected control (Table 2). These two treatments were in the same statistical group with healthy control. Root fresh weight was also significantly increased by all mycorrhizal species except *G. fasciculatum* and the highest effect was related to *F. mosseae*, *G. etunicatum* and *R. irregularis*, that increased this parameter by at least 53% compared to the infected control and these were placed in the same statistical group with healthy control. All endomycorrhizal treatments except *G. fasciculatum* significantly increased the root dry weight. Compared to the infected control, the highest effects were related to *F. mosseae*, *G. versiform* (both with 70% more root dry weight) and followed by *G. claroideum* (with 60% more root dry weight). These three treatments also significantly increased root dry weight compared to healthy control.

Colonization of common bean roots by AMF

In this study, all AMF were able to colonize common bean roots by more than 30% and there were significant differences between the colonization rates by different mycorrhizal species. *G. fasciculatum* and *A. longula* had the highest occurrence of root colonization (76 and 72%, respectively) and the lowest occurrence of root colonization was observed in *G. versiform* (31%) (Figure 2).

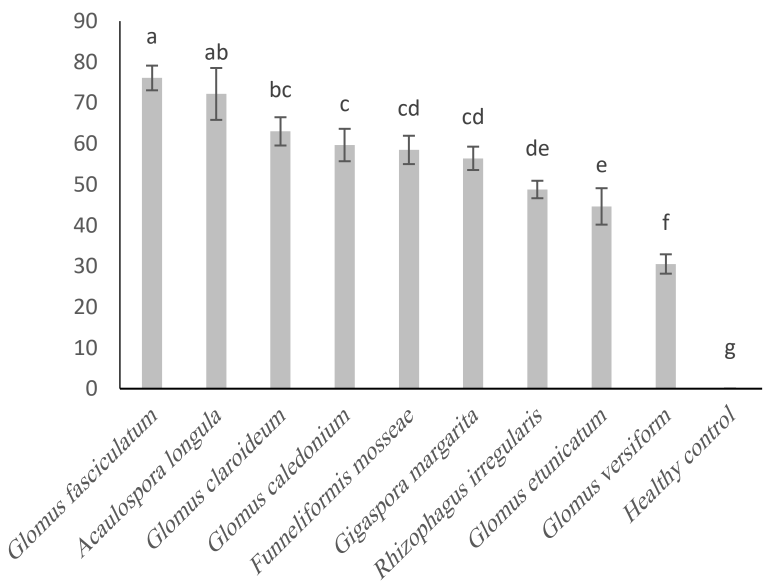

Figure 2. Colonization of common bean (*Phaseolus vulgaris* L.) roots by arbuscular mycorrhizal fungi. The means were compared using Duncan's multiple range test at 5% probability level. Bars with common letters are not significantly different.

Discussion

Rhizoctonia solani is a destructive fungal pathogen of common beans that can greatly reduce the yield and quality of this crop. Despite its effectiveness, the use of chemical fungicides against this pathogen is limited due to many associated risks (Marzouk *et al.* 2021). Beneficial microorganisms such as arbuscular mycorrhizal fungi are promising alternatives to replace or reduce the application of fungicides in agriculture (Alabouvette *et al.* 2006). In addition to their benefits for the management of plant diseases (Atwa 2018), AMF also contribute to the efficiency and sustainability of ecosystems (Gianinazzi *et al.* 2010). In the present study, nine different AMF species were inoculated to common bean plants to evaluate their effects in protecting the plants against *R. solani* and their effects on the growth parameters of common bean plants at the presence of the pathogen.

In this study, most of the mycorrhizal species decreased the damages of *R. solani* on common bean plants. This effect was evident from both the disease incidence and disease severity index in mycorrhizal plants; six out of the nine mycorrhizal species (*F. mosseae*, *G. margarita*, *G. claroideum*, *G. etunicatum*, *G. versiform* and *G. caledonium*) reduced the disease severity index and the first four these, also reduced the disease incidence. The effectiveness of some of these AMF species against *R. solani* has been previously reported in common bean or some other plants. For example, in cucumber plants, mycorrhization by *F. mosseae*, reduced the disease severity caused by *R. solani* from 65% (in non-mycorrhizal plants) to 21% (Aljawasim *et al.* 2020). Inoculation of potato seedlings by *G. etunicatum* enhanced the yield and the growth parameters (fresh and dry weight of roots and the number of tubers) and also reduced the disease severity by 60% -71.2% in shoots and crown (Yao *et al.* 2002). The effect of five AMF species namely *G. intraradices*, *G. fasciculatum*, *G. versiform*, *Glomus hoi* and *G. mosseae* has been studied against stem canker of potatoes caused by *R. solani*. These AMF species and their combination were able to decrease the disease (Mohammadi *et al.* 2020). Hafez *et al.* (2013) studied the effect of a mixture containing five AMF species including *F. mosseae*, *R. intraradices*, *G. clarum*, *G. margarita* and *Gigaspora gigantea* for reducing the severity and incidence of *R. solani* and enhancing the growth parameters in common bean and this mixture reduced the severity and incidence of the disease.

It has been stated that AMF mainly reduce the infection by various soil pathogens in plant roots and thus reduce the disease incidence and disease severity index (Bagy *et al.* 2019). Various mechanisms have been proposed by which AMF can increase the resistance of plants to soil-borne plant pathogens. Such mechanisms include competition with pathogens in rhizosphere, mycorrhizosphere and plant roots for infection sites and host photosynthetic products (Azcón-Aguilar & Barea 1996), increasing the resistance / tolerance of AMF-inoculated plants to the invasion of some plant pathogens due to their improving effect on plant nutrient status (Kareem & Hassan 2014), reducing or compensating root damages (Abdel-Fattah *et al.* 2011) due to the role that AMF play in absorption and transfer of nutrients to the roots, increasing the vegetative growth of plants by expanding the external mycelia and causing morphological changes in the roots (Bever *et al.* 2009; Liu *et al.* 2019), changes in the composition of the microbial communities of mycorrhizosphere (Singh *et al.* 2000), activation of plant defense mechanisms against soil pathogens (Miozzi *et al.* 2019; Shasmita *et al.* 2019) through biochemical, physiological and structural changes in the plant (Amer & Abou El 2008) which are thought to alter resistance responses to potential attackers both locally and systematically (Amer & Abou El 2008).

In the present study, variable results were observed for the effects of different AMF species on the incidence and severity of the disease caused by *R. solani* on common bean plants. *F. mosseae* and *G. claroideum* showed the highest inhibition of the disease and caused more than 40% reduction in the disease severity compared to the infected control. In contrast, three species namely *A. longula*, *G. fasciculatum* and *R. irregularis* did not have a significant effect on disease severity. This was in contrast with the results of some previous reports where *G. fasciculatum* and *R. irregularis* (syn: *R. intraradices*) reduced the damages of *R. solani* on potato seedlings (Mohammadi *et al.* 2020), application of *R. irregularis* in combination with Nitroxin biofertilizer caused a significant reduction in disease severity caused by *Fusarium oxysporum* f. sp. *ciceris* on chickpea (Ghorbani *et al.* 2022) and *R. intraradices* reduced the incidence and the disease severity of *R. solani* on common bean (Matloob & Juber 2013) and tomatoes (Amer & Abou El 2008). These differences may be explained by this fact that the biocontrol effect of AMF depends on several factors such as the species of the AM fungus involved, the substrate, the host plant and even its cultivar (Yao *et al.* 2002; Whipps 2004; Moarrefzadeh *et al.* 2022).

Despite the presence of the pathogen, different AMF studied in this work significantly compensated the damages from *R. solani* by enhancing the plant growth. All mycorrhizal treatments showed such enhancements in root length, fresh and dry weight of roots (except *G. fasciculatum*) and fresh weight of shoots (except *G. versiform*). Interestingly, the root dry weights of diseased plants in three AMF treatments (*F. mosseae*, *G. versiform* and *G. claroideum*) had increased to a level that was even significantly more than the healthy control. The positive effects of mycorrhizae on plant growth have been proven in many other studies (Matloob & Juber 2013; Matrood & Al-Taie 2017; Aljawasim *et al.* 2020; Mohammadi *et al.* 2020; Moarrefzadeh *et al.* 2021b). Increasing the growth of diseased plants by AMF plays a vital role in compensating for damage caused by plant pathogens (Wu *et al.* 2021). The three species of *G. fasciculatum*, *A. longula* and *R. irregularis* that did not reduce the severity of the disease, were able to reduce the adverse effect of the pathogen on infected plants, and this was determined by the improvements in some plant growth parameters at the presence of the pathogen. Improved plant growth may be due to the fact that mycorrhizal association with the plant greatly increases the uptake of water and nutrients by host plants by expanding its mycelial network in the rhizosphere. A better nutrition status increases chlorophyll content and photosynthetic efficiency in leaves, improves hormone homeostasis and ultimately leads to biomass accumulation, increased growth and yield and product quality (Chen *et al.* 2017; Shi *et al.* 2017; Aseel *et al.* 2019; Aslani *et al.* 2019). Biocontrol agents may also improve plant growth by restricting the growth of *R. solani* and preventing the disease (Matloob & Juber 2013).

In this study all studied AMF were able to colonize bean roots but there was significant differences in the colonization rates by different mycorrhizal species. Some species had high (more than 72% by *G. fasciculatum* and *A. longula*), and some had low (31% by *G. versiform*) colonization rates, but there was no relationship between the rate of root colonization by AMF species and disease severity; *G. fasciculatum* and *A. longula*, despite their high root colonization rate, had no significant effect in reducing the severity of the disease, while *G. versiform*, despite its low root colonization rate, was able to significantly reduce disease severity. Some other researches (St-Arnaud *et al.* 1994; Mark & Cassells 1996) have also indicated a lack of correlation between mycorrhizal root colonization rates with disease reduction in the host plant. Although AMF form a beneficial symbiotic relationship with the roots of most terrestrial plants (Shi *et al.* 2017), but the rate of colonization varies highly depending on the type of the host plant and AMF species (Chen *et al.* 2013; Chen *et al.* 2017). Dugassa *et al.* (1996) reported that the symbiotic effects of AMF on plant health, depends more on the host plant genotype and the pathogen than the rate of colonization by AMF.

The results of this research provide some evidence that AMF have high potential for the biological control of *R. solani* in common bean plants. Most of the studied AMF species reduced the severity of the disease and some also reduced the incidence of Rhizoctonia root rot on common bean plants, which indicates that AMF inoculation leads to protection and increased plant tolerance against the pathogen. In addition, treatments by all mycorrhizal species increased most of the growth parameters studied compared to the infected control. The beneficial effects of AMF varied depending on the species of the AMF; In plants inoculated with *G. mosseae* and *G. claroideum*, the disease severity was lower than other species and *G. mosseae* showed the most positive effect on plant growth. Therefore, inoculation with appropriate AMF species can help common bean plants to compensate the damages caused by *R. solani* and reduce the harmful effects of the pathogen. In summary, most of the mycorrhizal species used in this study can be considered as environmentally friendly, cost-effective and promising biocontrol options in the management of this soil-borne disease and as biofertilizers to stimulate the growth of common bean plants. Further research is needed to investigate the effect of these AMF species and also their combination(s) in controlling this pathogen in field conditions. The mechanisms by which AMF act as biological control agents and successful growth promoters also need more investigation.

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