

ORIGINAL PAPER

Potential of Arbuscular Mycorrhizal Fungi against Charcoal Rot of Sesame and Optimized Fertilization for Enhancing Growth, Productivity, and Nutrient Uptake

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ABSTRACT

This study aimed to evaluate the influence of arbuscular mycorrhizal fungi (AMF) (*Glomus mosseae* and *G. intraradices*) individually and combined with fertilization levels of superphosphate (SP) in rock phosphate form and potassium sulfate (PS) on sesame plant growth, productivity, and nutrient uptake. Results showed that AMF inoculation combined with different SP and PS rates significantly decreased damping-off (Pre and post emergence-damping off) and charcoal rot diseases compared with untreated control. Also, root colonization with AMF of sesame was high in the plants treated with SP at the low-level of AMF+SP (2.0), which was more and significantly effective for decreasing disease incidence gradually with increasing the fertility level of SP. The yield components of sesame plants treated with AMF in the presence of SP and RF levels were significantly increased compared to the control of non-treated plants. The results showed, also, that the AMF increased the nutrient uptake of PO₄, K, and Na in shoots and roots. AMF, also, enhanced the activity of enzymes involved in synthesizing sucrose, and essential sugars in plant metabolism, while reduced the activity of enzymes involved in the breakdown of sucrose. In addition, AMF increased levels of soluble sugars, proteins, and amino acid production in the plant. In conclusion, sesame cultivation in the presence of AMF with low SP and PS fertilizers can control damping-off and charcoal-rot in addition to improve plant growth, nutrient uptake, and crop parameters.

Keywords: *Sesamum indicum*, *Macrophomina phaseolina*, Mycorrhiza, Fertilizer rate diversity, Biochemical analysis.

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INTRODUCTION

Sesame (*Sesamum indicum* L.) is one of the richest sources of oil (46-52%), protein (18-20%) and different essential minerals such as calcium, phosphorus, and potassium, plus vitamins like vitamin E. The cultivated area of sesame in Egypt is about 70000 fed. and the production is of 3.18 t/f. (Anon. (FAOSTAT), 2021).

Sesame is infected with numerous soil borne fungi that have adverse effect on its seed quality and quantity of total seed production. In this concern, damping-off (DF) and charcoal-rot (CR) diseases caused by *Macrophomina*

phaseolina (Tassi) Goid are the most severe and destructive diseases causing seed yield losses in Egypt and the world (Khalifa, 2003). The worldwide sesame yield losses due to infection with CR reached 57% (Bashir *et al.*, 2017), whereas loss was about 5% or more in Egypt (Bedawy and Moharam, 2019). Applying the biotic agents such as mycorrhizal fungi protected the growing sesame plants from DF and CR diseases and significantly increased plant growth and seed yield. In this regard,

AMFs are important bioagents in sustainable agriculture, where they can enhance plant-water relations and influence the environment based on the used agrochemicals and high density in the field. Mycorrhiza is the symbiotic relationship between obligate soil-borne fungi and host plant roots (Rebeca *et al.*, 2013). The AMF can improve disease control, plant growth, increase mineral uptake, and reduce the use of fertilizers. Plant water status improvement and changes in water relations have been attributed to a variety of mechanisms, some of which are not directly linked to phosphorus sustenance or water uptake (Davies *et al.*, 1996). This symbiosis is more distinguished from others by its lack of host specificity. The AMFs are potential as a biofertilizer in which the fungus penetrates the cortical cells of a vascular plant's roots (Sullia, 1991). The AMFs initially penetrate root cells and develop into the root cortex of the host plant to generate two types of specialized structures, namely arbuscular and vesicles. Plant treatments with AMF are intended to lessen transplant stress and increasing soil hydration and fertility. Also, AMFs association can allow the treated plant host to obtain nutrients in an organic form that would otherwise be unavailable. Compared to the control of non-treated plants with AMF, mycorrhizal root structures can successfully take up phosphorus from lower amounts (Höweler *et al.*, 1981). In this regard, the AMFs is responsible for up to 80% of the total phosphorus uptake by treated plants (Marschner and Dell, 1994). An increase in host plant development, ascribed to a rise in nutrient uptake, is one of the most significant effects of AMF inoculation, especially in plants with low soil mobility and nutrient concentration. By absorbing the necessary carbon, providing the plant with nutrients, and increasing the efficiency of photosynthesis, they induce the chlorophyll organs of the plant to expand, and this demonstrated that mycorrhiza-inoculated maize plants have more dry matter than non-inoculated plants due to salinity. Additionally, compared to plants not inoculated with mycorrhiza, tomatoes under salt stress had more dry weight in their roots and shoots (Al-Karaki, 2006). Several studies have been reported on using plant growth regulators or AMF to lessen the negative impacts of environmental stress. The AMFs are distinct microorganisms that live in the rhizosphere and form symbiotic colonies with most plants. In addition to increasing

inorganic nutrients in plants, the AMFs can also increase plants' resistance to environmental stresses by enhancing the regulation of osmotic adjustment, stimulating growth regulators, and increasing photosynthesis (Rabie and Almadani, 2005). According to Allen (1991), the fungus *Glomus fasciculatum*, also, known as AMF, has a symbiotic relationship with plants and increases their nutrient content. During the past few decades, several studies on AMF have predicted their prevalence in a wide range of plant hosts, various environments, and diversity in quality and quantity (Augé, 2004; Ahmed *et al.* 2010; Bitterlich *et al.*, 2018 and Bedawy and Mohram, 2019).

A member of the Pedaliaceae family, the genus *Sesamum* contains the flowering plant known as sesame, which is cultivated for its highly nutritious edible oil seeds and is conventionally used for direct consumption and as a source of oil of excellent quality due to the presence of natural antioxidants like sesamin and sesamol, where it is widely naturalized in tropical regions through the world (Yamashita *et al.*, 2003). Due to its sub-marginal land cultivation, lack of high-yielding cultivars, and innate resistance to biotic and abiotic stress, sesame's yield potential is not outstanding, given the rising economic relevance of food, oil, and medicine. Sesame seed phospholipid cephalin has been found to have hemostatic properties. However, the oil has numerous uses in medicine and pharmaceuticals (Anilakumar *et al.*, 2010). Compared to the non-mycorrhizal treated plants, mycorrhizal plant growth characteristics such as plant length, number of roots, and number of leaves were examined and statistically assessed (Plenchette *et al.*, 1983). In this research, a study has been done to high light the effects of AMF on sesame plant growth. In addition to studying some biochemical changes in the plant after treatment, the shoot and root biomass and the biochemical elements' contents were evaluated. Also, this study aimed to control sesame damping-off and charcoal rot diseases using AMF and fertilization agents.

MATERIALS AND METHODS

Isolation, purification and identification of the causal pathogen of sesame damping-off and charcoal rot:

The causal pathogen was isolated from diseased sesame plant samples showing typical symptoms of damping-off and charcoal rot collected from different localities of Assiut

Governorate. After the purification of the obtained fungal isolates, the fungal isolates were identified according to their morphological characteristics as described by Domsch *et al.* (1980) and Sutton (1980) at Assiut University Mycological Centre, Assiut. The isolate demonstrated its high pathogenic ability, re-isolated, preserved on PDA slant agar in tubes, incubated for seven days at 27°C, and then kept at 4°C for further studies.

Isolation of arbuscular mycorrhizal fungi

Arbuscular mycorrhizal fungi were isolated from the rhizosphere of maize plants by wet sieving according to Gerdemann and Nicolson (1963). Both *Glomus mosseae* and *G. intraradices* were the most dominant species in the mycorrhizal inoculum.

$$\begin{aligned} \text{Damping-off(\%)} &= \frac{\text{Number of dead seedlings as pre and post emergence damping-off}}{\text{Total number of planted seeds}} \times 100 \\ \text{Charcoal rot} &= \frac{\text{Number of plants with charcoal rotted plants}}{\text{Total number of planted seeds}} \times 100 \end{aligned}$$

Effect of applying AMF individually and combined with levels of superphosphate (SP) and potassium sulfate (PS) fertilizations on the incidence of damping-off (DF) and charcoal-rot (CR) diseases, plant growth, and seed oil content of sesame:

A- Greenhouse experiment:

Plastic pots 35 cm in sterilized plastic pots containing formalin sterilized soil were artificially infested with the inoculum of the potent virulent isolate of *M. phaseolina*, causing sesame DF and CR diseases as described before. At the same time, mycorrhizae addition was done at the rate of 500 spores pot⁻¹ using a hemocytometer. Also, superphosphate (P₂O₅) was added to the soil before planting at rates 0.5, 1.0, 1.5, 2.0g and 5.0kg⁻¹ in the pots, and potassium sulfate (K₂O) was added at rates 0.25, 0.5, 0.75, 1.0g and 5.0kg⁻¹ in the pots. Only pots that received inoculum of *M. phaseolina* were served as a control. Three pots as replicates were used for each treatment in a completely randomized experimental design and six sterilized sesame seeds of cultivar Giza 32 were sown in each pot. Pots were irrigated when it was necessary and checked daily. Percentages of DF and CR incidence of sesame were calculated 21 and 90 days after sowing, respectively, using the two formulae mentioned before. Moreover, plant roots colonized by AMF were sampled during

Pathogenicity test:

The pathogenicity test was conducted for 15 isolates of *M. phaseolina* on sesame Giza 32 cultivar in the open greenhouse. Sesame seeds were surface sterilized with 1.0% sodium hypochlorite (SH) solution for 2 min and then washed three times with sterile distilled water (SDW). Then 35cm in diameter sterilized plastic pots containing formalin sterilized sandy loam soil were artificially infested with 3% inoculum of each tested fungal isolate then irrigated directly. The pots were then sown each with seven seeds, and four replicates (pots) were used for each tested fungal isolate. Percentages of sesame damping-off (DF) and charcoal rot (CR) were assessed 21 and 90 days after planting, respectively, according to the following two formulae:

sesame growing season and microscopically examined in the laboratory, and the presence of AMF spores was then confirmed.

B- Field experiments:

The experiments were conducted at the Experimental Farm of Al-Azhar University, Assiut Governorate, Egypt, during 2020 and 2021 growing seasons. The farm soil was sandy loam and naturally infested with *M. phaseolina*. The sowing date for both conducted field trails was the 1st of May. The sterilized seeds of Giza 32 sesame cv. were sown in hills at edges 50 cm apart and 3.5 cm long, and the distance between hills was 20 cm, and five seeds were sown in each hill. Each plot included six ridges, and the plot size was 10.5m² (3.5×3.0 m²) in a randomized complete block experimental design with three replicates (plots).

In this study, the AMF treatment was used individually and in combination with two fertilizers (superphosphate P₂O₅ and potassium sulfate K₂O) at four levels. The superphosphate was added before planting at 50, 100, 150, and 200 kg fed⁻¹ rates. These rates equal 125, 250, 375, and 500 g plot⁻¹ (10.5 m²). On the other hand, potassium sulfate fertilizer was added at 25, 50, 75, and 100 kg fed⁻¹. These rates were, also, equal to 62.5, 125, 187.5, and 250g plot⁻¹ (10.5 m²). After 21 days of sowing, sesame plants were thinned into one plant per hill. The

recommended cultural practices for sesame production were followed throughout the growing season.

Percentages of CR incidence of sesame was calculated 90 days after sowing as mentioned before. At harvest time, 110 days after sowing, samples of 10 apparently healthy plants were randomly collected from each plot to estimate some crop parameters *i.e.* plant height (cm), number of capsules per plant, seed yield per plant (g), weight of 1000-seed (g), and percentage of seed oil content.

Percentage of seed oil content:

In this study, 15 g of dried sesame seeds collected from plants of each treatment, grown under greenhouse experimental conditions, were used to estimate the oil content of seeds using Soxhlet apparatus and petroleum ether (BP 40-60°C) as solvent solution according to the official standard method described by Anon. (1995), and then the oil content was expressed in percentage.

Plant root and shoot tissues extraction:

In this study, the known weight of the oven-dry plant matter (shoots or roots) obtained from the treatments tested under greenhouse experimental conditions was extracted at the known volume of water (w/v) at 90°C water bath. The extracts were then preserved in a refrigerator to studying some biochemical changes in the plant after treatment during the following experiments.

1- Determination of soluble metabolites:

According to the methods described by Lowry *et al.* (1951), Dubois *et al.* (1956), and Lee and Takahashi (1966), the soluble proteins, total free amino acids, soluble sugars, and hydrolyzable sugars were determined calorimetrically in the previously prepared extracts. The content of each referred metabolite was expressed in mg g⁻¹ dry weight of the studied plant.

2- Minerals analysis:

In this study, the phosphate (PO₄⁻³) was estimated based on forming a molybdate blue color with ascorbic acid as a reducing agent according to the method described by Vogler (1965). At 720 nm, the extinction of the molybdate blue color was measured. Also, sodium and potassium (Na⁺ and K⁺) were determined by the flame emission technique (Carl-Zeiss DR LANGE M 7 D flame photometer), which is considered a rapid and sensitive method for determining sodium and potassium (Williams and Twine, 1960).

Statistical analysis:

All data obtained were statistically analyzed by the MSTAT-C program version 2.10. Duncan's multiple range tests for means of tested treatments comparing and the least significant difference (L.S.D.) at the $P \leq 0.05$ probability level was used as described by Gomez and Gomez (1984).

RESULTS

Pathogenicity test of the isolated *M. phaseolina* isolates:

Data in Table (1) show that all the tested 15 *M. phaseolina* isolates caused DF and CR diseases associated with discoloration of infected tissues with different disease incidence degrees.

Table (1): Pathogenicity test of various *M. phaseolina* isolates determined on Giza 32 sesame cultivar under greenhouse conditions.

| Isolates NO. | %, Damping off | %, Charcoal-rot |
|--------------|----------------|-----------------|
| 1 | 7.14 | 24.99 |
| 2 | 14.28 | 60.69 |
| 3 | 24.99 | 21.41 |
| 4 | 42.84 | 46.41 |
| 5 | 21.42 | 74.97 |
| 6 | 28.56 | 49.98 |
| 7 | 17.85 | 67.83 |
| 8 | 32.13 | 28.56 |
| 9 | 3.57 | 17.85 |
| 10 | 21.42 | 53.55 |
| 11 | 10.71 | 39.27 |
| 12 | 14.28 | 14.28 |
| 13 | 10.71 | 64.26 |
| 14 | 28.56 | 32.14 |
| 15 | 7.14 | 10.71 |
| Control | 0.00 | 0.00 |
| LSD at 0.05 | 14.40 | 16.23 |

The obtained results showed that isolate No.5 recorded the highest percentage of CR infection, being 74.97%, followed by isolates No. 7, 13, and 2, being 67.83, 64.26, and

60.69% without significant differences respectively, while isolates No.10 and 6 came next and caused 53.55 and 49.98% CR, respectively. In contrast, isolate No. 15 recorded the lowest CR infection, being 10.71%, followed by isolates No. 12, 9, and 3, being 14.28, 17.85, and 21.41%, respectively.

Effect of applying AMF individually and combined with levels of SP and PS fertilizations on the incidence of DF and CR diseases, growth, and seed oil content of sesame:

A- Greenhouse experiment:

Data shown in Table (2) reveal that when AMF inoculated alone or in combinations with different rates of SP and PS significant decrease in sesame DF and CR diseases was occurred compared with untreated control. However, results of the interactions between AMF with different treatments by fertilizers showed different responses. In this regard the (AM+SP 2.0 g) treatment was more effective and significantly decreased DF, being 5.55% and no CR was occurred. In addition, the rest of the treatments exhibited the highest reduction of both DF and CR, being 11.10&5.55%, 16.65&5.55%, and 21.20&11.10, respectively. Also, rates of PS with AMF treatments were of significant effect for decreasing the percentage of CR disease incidence and decreasing DF, with exception for treatment AM+PS (0.25), which recorded 38.85% DF.

Table (2): Effect of mycorrhizae combined with SP and PS fertilizations on the incidence of sesame DF and CR diseases under greenhouse conditions.

| Treatments | %, Damping-off | %, Charcoal-rot |
|-------------------------|----------------|-----------------|
| Control (infested soil) | 38.85 | 61.05 |
| AM | 22.20 | 11.10 |
| AM +SP (2.0 g) | 5.55 | 0.00 |
| AM +SP (1.5 g) | 11.10 | 5.55 |
| AM +SP (0.1 g) | 16.65 | 5.55 |
| AM +SP (0.5 g) | 22.20 | 11.10 |
| AM +PS (1.0 g) | 22.20 | 11.10 |
| AM +PS (0.75 g) | 33.30 | 22.20 |
| AM +PS (0.5 g) | 16.65 | 16.65 |
| AM +PS (0.25 g) | 38.85 | 27.75 |
| LSD at 0.05 | 17.39 | 14.65 |

B- Field experiments:

Under field conditions, all evaluated treatments reduced the incidence of sesame CR compared to the control, as shown in Table (3). Results, also, showed that treatments AM+SP at 200 Kg and AM+SP at 150 kg had the highest activity and significantly reduced the percentages of sesame CR compared to the control. The second best treatment was AM+SP at 100 kg. However, the lowest reduction in the incidence of CR was resulted from combined treatment of AM+PS at 25 Kg. The activity of these treatments was significantly increased by increasing the concentrations of both evaluated fertilizers. Moreover, the plant roots colonized by AMF were increased during the sesame growing season following the microscopic examination, and the presence of AMF spores was also confirmed.

Effect of applying AMF individually and combined with levels of SP and PS fertilizers on sesame plant growth characters and seed oil content:

Results in Table (4) indicate that applying mycorrhiza singly or combined with fertilization treatments significantly increased plant height (cm), the number of capsules per plant, seed yield per plant (g), the weight of 1000-seed (g), and seed oil content (%). Combined treatments were generally better than single treatments in the two growing seasons. Values of sesame plant parameters were significantly increased with the dual inoculation of mycorrhizae and combined with levels of SP and PS fertilizers. The combined treatment of AM+SP (200 kg) caused higher values of growth parameters than the singly AM-treated plants and the rest of the transactions.

Concerning the seed oil content of sesame, there were statistical differences between all treatments and the control. The seed oil content of sesame was significantly influenced by using AM in combinations with different SP and PS fertilizers application rates. Increasing the fertilizers level increased the oil content significantly. The maximum seed oil contents, being 60.0 and 58.93% were occurred due to using AM+SP (200 kg) and AM+SP (150 kg) treatments, which were significantly superior over other sources of fertilizer treatments in both experimental seasons, followed by AM treatment. In contrast, control treatment yielded the lowest seed oil content.

Table (3):Effect of mycorrhizae combined with SP and PS fertilizations on sesame CR disease incidence under field conditions. Mean over the 2020 and 2021 growing seasons.

| Treatments | %, Charcoal-rot during | |
|----------------|------------------------|-------|
| | 2020 | 2021 |
| Control | 75.27 | 74.49 |
| AM | 5.00 | 5.00 |
| AM+SP (200Kg) | 3.02 | 0.00 |
| AM+SP (150kg) | 4.10 | 0.00 |
| AM+SP (100 kg) | 5.18 | 5.33 |
| AM+SP (50 kg) | 6.33 | 6.00 |
| AM+PS (100Kg) | 5.10 | 5.16 |
| AM+PS (75 Kg) | 7.54 | 7.93 |
| AM+PS (50 Kg) | 5.66 | 5.99 |
| AM+PS (25 Kg) | 9.53 | 8.65 |
| LSD at 0.05 | 0.97 | 0.93 |

Effect of applying AMF individually and in combination with levels of SP and PS fertilizers on the major biochemical metabolites content of sesame under greenhouse conditions:

Soluble sugars (SS) are the most cell metabolites that reflect the plant's health; they are considered the first product of photosynthesis. Generally, most SS accumulation was in sesame shoots (about 4 to 5 folds of those in roots). The highest SS accumulation was observed in plants treated with Mycorrhiza (AM) and AM + at 200 Kg SP. The soluble sugar content ranged between 33.21- 60.04mg g⁻¹ DW in the shoot and 5.50 - 2.20 in roots during both seasons 2020 and 2021 (Table, 5). In both growing seasons, values at AM treatment were significantly higher than other tested treatments of fertilizer sources, followed by AM with 150 and 200 Kg SP. Also, the same improvement effect was produced when PS fertilizer at 100 Kg was added with AM.

On the other hand, control treatment showed a low-level value of soluble sugar in both experimental growing seasons. Soluble sugar content expressed in the consumer acceptance test the higher the sugar content, the better the acceptance. The experimental finding reveals that the sugar content was significantly higher due to using AM treatment than in control or

with different SP and PS fertilizers rates. It expressed that AM treatment applied at the total dose could gain the increasing sugar content as compared to the control treatment.

Plant height values at the harvest stage were increased with an increase in AM fungi and is inoculated in combinations with different fertilizers levels.

This study illustrated that the free amino acids (FAA) contents of sesame shoots (resembling about 2-folds of roots FAA) were significantly increased by the effect of AM fungi and inoculated in combinations with different rates of superphosphate and potassium sulfate compared with the control. The most notable rise in FAA contents was under the AM + K at 50 Kg and AM + K at 25 Kg.

Soluble proteins (SP) approximately followed the same trend as FAA by recording the highest production under AM treatments against control. Thus, these treatments, especially PS at 25 and 50 Kg, were better for treating the soil to improve sesame production of FAA and SP.

Plant nutrient ions e.g., PO₄, K, and Na uptake in roots and accumulation in shoots were influenced by Mycorrhizal fungi and those inoculated in combinations with different SP and PS fertilizers application rates (Table, 6). Concerning the total potassium content, the obtained data showed a significant rise in shoots and roots compared to the control treatment. The potassium contents ranged between 11.49 to 13.07 mg g⁻¹ DW and 1.93 to 1.78 mg. g⁻¹ DW, respectively. The results indicated that using AM combined with the concentration of SP at 50 and 100 Kg gave the highest value of potassium in plant shoots. Thus, shoots potassium contents were generally higher in treated plants than in control. The formerly recorded peaks with AM + 50 and 100 Kg potassium fertilizers.

Phosphates were produced and accumulated in shoots with about 20 times more than those in roots, and they were highly accumulated in plants grown in AM-treated soils (especially those coupled with 50 Kg superphosphate and 100 Kg potassium sulfate). Phosphate means fluctuated between 19.67 to 9.72 mg g⁻¹ DW and 1.61 to 0.04 mg g⁻¹ DW in shoots and roots, respectively.

Data of Na content in Table (6) behave similarly to total phosphate. Otherwise, the opposite was obtained with potassium contents. Sodium means in shoots, and roots ranged between 3.37 to 1.67 mg g⁻¹ DW and 2.57 to

0.06 mg g⁻¹ DW, respectively. The highest mean of Na⁺ content in shoots and roots was recorded in mycorrhiza-plants associations coupled with 200 Kg super phosphate. The lowest value shown of Na⁺ content in shoots and roots was recorded in sesame plants treated with AM+ PS at 25 Kg.

The results in Table (6) also show that arbuscular mycorrhizal fungi (AMF) increased the nutrient uptake of (PO₄, K, and Na in shoots and roots with low doses of fertilizers). Moreover, with the continued increase in levels of SP fertilizer, the nutrient uptake of these elements was reduced compared to non-Mycorrhizal and control treatments.

DISCUSSION

The pathogenic ability of the isolated fungi was tested under greenhouse conditions, and all the *M. phaseolina* isolates obtained caused sesame damping-off (DF) and charcoal rot (CR) association with discoloration of infected sesame plant tissues. These results are in agreement with those reported by Plenchette *et al.* (1983); El-Fiki, *et al.* (2002); Khalifa (2003); Ahmed *et al.* (2010), and Bedawy and Moharam (2019). In this study, under greenhouse conditions, planting sesame seeds in soil inoculated with AM alone or in combinations with different SP and PS rates significantly decreased DF and CR diseases in most treatments compared with the control. Khalifa, (1997) and El-Fiki, *et al.* (2004) reported that AM fungi play an important role in reducing charcoal rot of sesame. Inoculating plants with AM can potentially increase or maintain yields and allow for reducing fertilizer application (Douds *et al.*, 2008). Chitra (2015) presented a study that established the association of AM fungi with sesame-enhanced growth when compared with the control. Also, Khalifa *et al.* (2012) reported that calcium and AM play an important role in reducing many plant diseases. The positive effect may be due to the role of calcium in the plant *i.e.*, it has a critical metabolic role.

Under field conditions and natural infestation with *M. phaseolina* in each growing season of 2020 and 2021, all treatments reduced the incidence of sesame CR compared to the control. This result is in agreement with the results obtained by Castillo *et al.* (2012), who found that treating wheat, barley, and oats with AM fungi significantly increased their root

biomass. The AM infection has been reported to increase the uptake of nutrients by the roots

and the concentration of nutrients in the plant tissues. Thus, it can be concluded that the high rate of chemical phosphorus fertilizers application leads to antagonistic interaction with mycorrhiza. Mahmoud *et al.* (2008) revealed that inoculation with Rhizobium and VA-mycorrhizae under different sources of phosphorus fertilizer reduced peanut root rot disease and enhanced peanut yield. Khalifa *et al.* (2010) stated that VA mycorrhiza and some bio-fertilizers and different sources of mineral phosphorus were effective in controlling pod rot diseases and aflatoxin contamination in seeds of peanut

Mycorrhiza significantly increased root colonization in low-chemical phosphorus fertilizers (Soleimanzade *et al.* 2010). VA mycorrhizal fungi protect plants by reducing pathogenic activity and enhancing resistance due to the increase of antifungal chitinase enzymes in roots (Dehne *et al.* 1978). The AM application, also, increases lignin content in the root system (Ziedan *et al.* 2010). Also, values of growth parameters of sesame plants significantly increased yield components *i.e.*, the height of plant (cm), the number of capsules per plant, seed yield per plant (g), weight of 1000-Seed (g), and oil content (%) by inoculation of mycorrhizae combined with fertilization treatments. These results are in agreement with Ziedan *et al.* (2011), who reported that biological fertilizers recorded higher seed yields than NPK fertilizers. Also, Tohidi-Moghaddam *et al.* (2004) reported that phosphorous-solubilizing microorganisms increase the available phosphorous in the soil, which could improve the seed number in plants. Using mixed inoculum of mycorrhizal symbionts and biocontrol agents can be more effective than using a single species. The seed oil content was significantly influenced by AM fungi when inoculated in combinations with different rates of SP and PS fertilizers, and there were statistical differences between all treatments and the control.

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inoculum of mycorrhizal symbionts and biocontrol agents can be more effective than using a single species. The seed oil content was significantly influenced by AM fungus when inoculated in combinations with different rates of SP and PS of fertilizers, and there were significant differences between all treatments and the control. These results are in agreement with those reported by Ziedan *et al.* (2011) and Mahrous *et al.* (2015). The lowest seed oil content was obtained in the control treatment. This result could, also, be attributed to AM fungus's influence and inoculation in combination, which result in rapid change of nitrogen to crude protein in the seed and finally to oil.

Application of different AM fungi and in combinations with different rates of SP and PS fertilizers have shown significant enhancement in all yield parameters than the control for the growth and yield components measured in terms of plant height, the number of capsules and seeds per capsule, in both the growing seasons. Concerning the effects of mycorrhizae in combinations with different rates of SP and PS on soluble sugars (SS) accumulation were observed in plants treated with AM+200 Kg SP, and this result was also evinced in the studies of Dubey and Khan (1993). This study demonstrated that sesame shoots' FAA contents resembling about 2-folds of roots from FAA. Our results are in agreement well with those of previous studies reporting that free amino acids can be affected by numerous factors, including process techniques, environmental stresses, and agronomic conditions (Umata and Dabalo 2017). Soluble proteins followed the same trend as FAA by recording the highest production under AM treatments compared with the control. Thus these, treatments, especially, potassium sulfate, might be better for treating the soil to improve sesame production from FAA and SP.

The AMF positively impacts plant growth by forming symbiotic associations with plant roots. This symbiosis can increase nutrient uptake, improve water relations, and enhance plant stress tolerance. Also, AMF can influence plant physiology by altering the levels of certain compounds, such as soluble sugars, free amino acids, and soluble proteins (Bitterlich *et al.*, 2018). The increase in soluble sugars in plants treated with AMF is thought to be due to various factors, including improved photosynthetic capacity and nutrient uptake, as well as the breakdown of complex carbohydrates in the soil. Similarly, the increase in free amino acids and soluble proteins is likely due to increased nitrogen uptake and the breakdown of proteins in the soil and plant (Augé, 2004). AMF can, also, affect the balance among different metabolic pathways in the

plant. For instance, AMF can enhance the activity of enzymes involved in synthesizing sucrose, an essential sugar in plant metabolism, while reducing the activity of enzymes involved in the breakdown of sucrose. This effect can result in increased levels of soluble sugars in the plant.

Mycorrhizal fungi can directly affect root morphology, hydraulic conductivity, and the expression of plant aquaporins, all of which can affect plant water uptake. In addition to these direct effects, downstream effects may contribute to the observed increase in water and nutrient uptake and drought tolerance in plants associated with AMF. Mycorrhizal roots' improved water uptake efficiency and lower resistance to water flow in the substrate may be necessary for the frequently observed higher rates of soil drying in mycorrhizal systems (Ruiz-Lozano *et al.*, 2016).

The increase in soluble sugars in plants treated with AMF is thought to be due to several factors. First, AMF can enhance the photosynthetic capacity of plants, which can result in increase production of sugars. Second, AMF can improve the efficiency of nutrient uptake, particularly phosphorus, which can increase the plant's energy availability (Moradtalab *et al.*, 2019). This increase in energy availability can, also, result in increasing sugar production. Additionally, AMF can produce enzymes that break down complex carbohydrates in the soil, making them more available to the plant as an energy source.

The increase in free amino acids in plants treated with AMF is, also, likely due to a combination of factors. First, AMF can enhance nitrogen uptake from the soil, resulting in increasing amino acid production (Hajiboland *et al.*, 2015). Second, AMF can produce enzymes that break down proteins in the soil, making them more available to the plant. Finally, AMF can, also, stimulate the production of enzymes in the plant that break down proteins and other nitrogen-containing compounds, which can result in increasing amino acid production (Calvo-Polanco *et al.*, 2016).

Also, the increase in soluble proteins in plants treated with AMF is likely due to a combination of factors. First, AMF can increase the efficiency of nutrient uptake, particularly nitrogen, which can increase protein production. Second, the AMF can produce enzymes that break down proteins in the soil, making them more available to the plant. Finally, AMF can, also, stimulate the production of enzymes in the plant that synthesize proteins, resulting in increasing protein production.

Table (4): Effect of mycorrhizae combined with SP and PS fertilizers on some plant growth parameters of sesame during 2020 and 2021 growing seasons. Mean over of 2020 and 2021 growing seasons.

| Treatments | Plant height (cm) during | | Number of capsules per plant during | | Seed yield per plant (g) during | | 1000-Seed weight (g) during | | Oil seed content (%) during | |
|----------------|--------------------------|--------|-------------------------------------|--------|---------------------------------|-------|-----------------------------|------|-----------------------------|-------|
| | 2020 | 2021 | 2020 | 2021 | 2020 | 2021 | 2020 | 2021 | 2020 | 2021 |
| Control | 128.35 | 130.33 | 97.00 | 96.00 | 9.11 | 9.15 | 2.94 | 2.98 | 51.81 | 50.84 |
| AM | 176.33 | 177.00 | 180.50 | 181.33 | 16.86 | 17.41 | 4.93 | 5.00 | 57.18 | 57.93 |
| AM+SP (200Kg) | 183.00 | 182.33 | 195.00 | 196.00 | 22.35 | 22.80 | 5.04 | 5.91 | 60.10 | 60.00 |
| AM+SP (150kg) | 181.33 | 180.00 | 190.40 | 189.66 | 17.71 | 18.26 | 4.44 | 4.62 | 59.35 | 58.93 |
| AM+SP (100 kg) | 175.66 | 176.00 | 136.60 | 134.33 | 13.50 | 13.75 | 4.46 | 4.51 | 54.91 | 54.51 |
| AM+SP (50 kg) | 171.33 | 173.66 | 124.80 | 123.00 | 11.21 | 11.84 | 3.94 | 4.10 | 53.14 | 52.51 |
| AM+PS (100Kg) | 177.33 | 177.66 | 186.00 | 185.00 | 13.84 | 14.35 | 4.48 | 4.64 | 55.32 | 55.12 |
| AM+PS (75 Kg) | 160.33 | 162.00 | 140.60 | 138.00 | 12.34 | 12.46 | 4.10 | 4.00 | 54.31 | 54.00 |
| AM+PS (50 Kg) | 180.33 | 180.66 | 147.10 | 145.00 | 15.11 | 15.72 | 4.63 | 4.66 | 57.12 | 56.33 |
| AM+PS (25 Kg) | 160.21 | 161.00 | 116.50 | 116.00 | 10.48 | 10.43 | 3.89 | 3.93 | 52.51 | 51.88 |
| LSD at 0.05 | 7.97 | 8.34 | 5.31 | 6.27 | 1.07 | 1.51 | 0.90 | 0.97 | 1.24 | 1.84 |

The AMF increased the nutrient uptake of PO₄, K, and Na in shoots and roots with low fertilizers. These results are in agreement with the finding of Nafady *et al.* (2018), who studied the effect of AMF and phosphorus fertilizers, including SP, on wheat (*Triticum aestivum* L.) plants' growth, productivity, and nutrient uptake. The AM-inoculated wheat plants were significantly higher in growth, productivity parameters, and nutrient uptake than non-inoculated plants at all phosphate fertilizers levels.

The positive impact of AMF on host plant growth during any stress is often attributed to improved phosphorus nutrition, which was likely the case with the plants in our study (Johnson and Hummel, 1985). Under normal conditions, higher plants produce reactive oxygen species (ROS) in chloroplasts, mitochondria, and peroxisomes, and the production and removal of ROS are tightly regulated (Apel and Hirt, 2004). However, when plants are exposed to stress, this balance is disrupted, leading to oxidative damage to proteins, DNA, and lipids. The malondialdehyde (MDA) is a byproduct of this damage and can indicate the degree of membrane lipid peroxidation. Therefore, AMF can reduce ROS accumulation and prevent oxidative damage in plants subjected to environmental stresses.

Overall, the exact mechanisms by which AMF increases the levels of soluble sugars, free amino acids, and soluble proteins in plants are not fully understood and likely involve complex interactions between the fungus and the plant. However, the symbiotic association between AMF and plants can result in significant changes in plant physiology, including alterations in the levels of various compounds, ultimately resulting in improved plant growth and health

CONCLUSION

The present study suggested that the combined application of AMF (*Glomus mosseae* and *G. intraradices*) and low levels of phosphorus and potassium sulfate fertilizers of sesame plants is more effective than single treatment, where there was a significant improvement in plant growth, biomass and yield components. Furthermore, nutrient content of sesame can be increased as a result of mycorrhizal treatments. The results also indicated that the use of high fertilizer concentrations reduce the beneficial mycorrhizal effect on plant growth, yield production and nutrient uptake. Therefore, treatment with high levels of fertilizers is not recommended because they reduce the beneficial mycorrhizae of plant.

Table (5):Effect of mycorrhizae combined with SP and PS fertilizers on some soluble metabolites of shoots and roots of sesame plant under greenhouse conditions

| Content | Treatments | Soluble metabolites in shoots during | | Soluble metabolites in roots during | |
|------------------|----------------|--------------------------------------|------------|-------------------------------------|------------|
| | | 2020 | 2021 | 2020 | 2021 |
| Soluble sugars | AM | 60.04±10.24 | 66.09±2.82 | 5.5±0.79 | 7.2±0.53 |
| | AM+SP (2.0 g) | 40.22±2.37 | 68.21±3.68 | 2.2±0.05 | 3.02±0.25 |
| | AM+SP (1.5 g) | 49.83±3.12 | 73.72±9.19 | 3.72±0.37 | 2.72±0.49 |
| | AM+SP (0.1 g) | 50±3.1 | 57.42±5.87 | 13.56±10.02 | 4.68±1.18 |
| | AM+SP (0.5 g) | 45.55±1.76 | 56.55±5.95 | 3.02±0.7 | 3.02±0.05 |
| | AM+PS (1.0 g) | 41.37±3.41 | 39.84±2.91 | 7.98±1.96 | 3.87±1.2 |
| | AM+PS (0.75 g) | 56.23±4.85 | 22.58±9.54 | 4.6±0.52 | 2.61±0.43 |
| | AM+PS (0.5 g) | 39.96±1.18 | 31.2±5.08 | 6.98±1.03 | 3.7±0.27 |
| | AM+PS (0.25 g) | 34.09±3.51 | 37.29±0.16 | 2.2±0.69 | 1.34±0.46 |
| | Control | 33.21±2.2 | 37.8±1.41 | 4.98±0.55 | 4.14±0.23 |
| LSD at 0.05 | | 12.53 | 16.24 | 9.2 | 1.85 |
| Free amino acids | AM | 18.38±1.29 | 16.8±0.9 | 11.08±1.36 | 9.84±2 |
| | AM+SP (2.0 g) | 18.5±1.14 | 14.64±1.27 | 6.8±0.61 | 8.7±0.22 |
| | AM+SP (1.5 g) | 13.33±1.48 | 16.4±0.63 | 18.82±1.76 | 11.73±3.71 |
| | AM+SP (0.1 g) | 17.03±1.12 | 18.39±1.11 | 19.7±1.72 | 9.1±0.22 |
| | AM+SP (0.5 g) | 23.8±6.99 | 26.62±2.62 | 22.52±1.56 | 7.27±0.08 |
| | AM+PS (1.0 g) | 16.76±2.19 | 17.34±0.5 | 18.75±4.65 | 10.12±0.16 |
| | AM+PS (0.75 g) | 13.31±1.05 | 21.32±0.27 | 20.77±3.51 | 10.4±0.41 |
| | AM+PS (0.5 g) | 15.32±1.31 | 11.22±1.33 | 25.16±2.07 | 6.86±0.28 |
| | AM+PS (0.25 g) | 25.11±3.52 | 20.99±1.93 | 7.05±0.11 | 7.14±0.59 |
| | Control | 30.75±5.11 | 18.87±1.47 | 8.33±1.23 | 8.06±0.24 |
| LSD at 0.05 | | 9.36 | 4.05 | 10.25 | 4.02 |
| Soluble proteins | AM | 48.81±10.43 | 45.8±2.36 | 9.32±0.96 | 41.71±0.5 |
| | AM+SP (2.0 g) | 43.07±2.19 | 25.91±0.06 | 5.88±0.19 | 38.81±0.81 |
| | AM+SP (1.5 g) | 29.46±5 | 59.23±2.67 | 10.9±3.58 | 41.82±2.42 |
| | AM+SP (0.1 g) | 36.26±1.8 | 54.5±2.05 | 10.32±0.66 | 37.73±1.18 |
| | AM+SP (0.5 g) | 23.08±3.59 | 53.54±2.61 | 13.48±0.31 | 7.85±0.81 |
| | AM+PS (1.0 g) | 27.59±2.81 | 66.87±0.62 | 17.63±2.8 | 10.32±2.48 |
| | AM+PS (0.75 g) | 39.78±5.91 | 60.85±0.5 | 10.25±1.09 | 11.18±0.99 |
| | AM+PS (0.5 g) | 26.73±1.72 | 56.76±5.71 | 12.26±2.58 | 3.55±0.06 |
| | AM+PS (0.25 g) | 24.44±0.19 | 62.14±0.12 | 10.25±2.49 | 6.45±1.24 |
| | Control | 32.18±0.94 | 57.94±3.17 | 10.11±1.1 | 8.06±0.93 |
| LSD at 0.05 | | 12.92 | 7.63 | 5.5 | 4.0 |

Table 6. Effect of mycorrhizae combined with fertilization (superphosphate and potassium sulfate) on soluble ions.

| Content | Treatments | Soluble ions of shoots during | | Soluble ions of roots during | |
|-------------|----------------|-------------------------------|------------|------------------------------|-----------|
| | | 2020 | 2021 | 2020 | 2021 |
| Sodium | AM | 3±0.06 | 1.88±0.17 | 1.13±0.03 | 1.1±0.06 |
| | AM+SP (2.0 g) | 3.37±0.27 | 2.97±0.09 | 1.5±0.29 | 1.97±0.09 |
| | AM+SP (1.5 g) | 2.67±0.44 | 3.47±0.03 | 1.53±0.28 | 1.4±0.15 |
| | AM+SP (0.1 g) | 2.47±0.32 | 3.28±0.12 | 1.5±0.31 | 1.68±0.09 |
| | AM+SP (0.5 g) | 3±0.06 | 2.9±0.06 | 1.37±0.22 | 1.5±0.06 |
| | AM+PS (1.0 g) | 1.67±0.24 | 2.83±0.09 | 1.04±0.07 | 1.6±0.06 |
| | AM+PS (0.75 g) | 2±0.46 | 3.07±0.03 | 2.57±0.23 | 2.25±0.14 |
| | AM+PS (0.5 g) | 2.3±0.4 | 3.5±0.06 | 0.93±0.09 | 2.25±0.14 |
| | AM+PS (0.25 g) | 1.67±0.17 | 2.26±0.32 | 1±0.06 | 2.07±0.09 |
| Control | | 1.67±0.17 | 2.47±0.2 | 1.02±0.09 | 1.5±0.12 |
| LSD at 0.05 | | 0.86 | 0.43 | 0.59 | 0.31 |
| Potassium | AM | 11.49±1.84 | 9.33±0.12 | 1.93±0.49 | 1.94±0.3 |
| | AM+SP (2.0 g) | 14.47±0.83 | 11.57±0.37 | 1.53±0.12 | 1.06±0.08 |
| | AM+SP (1.5 g) | 13.97±1.5 | 9.6±0.85 | 1.02±0.15 | 0.75±0.03 |
| | AM+SP (0.1 g) | 14.8±1.75 | 11±1.23 | 1.53±0.22 | 1.15±0.03 |
| | AM+SP (0.5 g) | 13.37±0.75 | 17.5±0.17 | 1.32±0.41 | 1.23±0.19 |
| | AM+PS (1.0 g) | 13.3±0.2 | 12.63±0.64 | 3.02±1.01 | 1.45±0.16 |
| | AM+PS (0.75 g) | 11.97±1.32 | 14.07±0.58 | 1.33±0.22 | 0.7±0.1 |
| | AM+PS (0.5 g) | 14.43±1.87 | 8.46±1.12 | 2.28±0.19 | 1.06±0.04 |
| | AM+PS (0.25 g) | 17.27±1.22 | 14.57±0.21 | 1.67±0.35 | 0.15±0.01 |
| Control | | 13.07±1.63 | 13.17±0.37 | 1.78±0.24 | 0.11±0.01 |
| LSD at 0.05 | | 3.97 | 2.00 | 1.21 | 0.39 |
| Phosphates | AM | 9.81±0.61 | 14.88±1.49 | 0.06±0.02 | 0.04±0.01 |
| | AM+SP (2.0 g) | 19.67±2.34 | 20.73±2.46 | 0.6±0.01 | 0.04±0.01 |
| | AM+SP (1.5 g) | 14.39±0.68 | 2.67±0.18 | 0.41±0.05 | 0.16±0.01 |
| | AM+SP (0.1 g) | 9.72±0.18 | 5.34±1.21 | 0.95±0.16 | 0.1±0.01 |
| | AM+SP (0.5 g) | 10.59±0.64 | 8.44±0.61 | 0.74±0.12 | 0.1±0.02 |
| | AM+PS (1.0 g) | 11.67±0.33 | 8.98±0.92 | 1.61±0.42 | 0.1±0.01 |
| | AM+PS (0.75 g) | 13.19±1.01 | 12.33±0.74 | 0.79±0.18 | 0.04±0.01 |
| | AM+PS (0.5 g) | 10.57±0.68 | 1.48±0.05 | 1.45±0.05 | 0.11±0.01 |
| | AM+PS (0.25 g) | 11.35±1.11 | 7.19±0.77 | 0.62±0.06 | 0.04±0.01 |
| Control | | 6.37±0.54 | 7.92±1.12 | 0.25±0.06 | 0.07±0.01 |
| LSD at 0.05 | | 2.93 | 3.41 | 0.46 | 0.03 |

CONFLICTS OF INTEREST:

The authors declare that they do not have any actual or potential conflict of interest

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