Guava Plants Growth and Soil Microbial Respiration in Response to Arbuscular Mycorrhizal Species Inoculation Under Alkaline Soil Conditions

Hoda A. Mahmoud[1]; Yasser T. A. Moustafa[2]; Walid F. A. Mosa[3]; R. M. El Adly[1] and Abou El Seoud I. I[1]

[1]Department of Soil and Agricultural Chemistry, Faculty of Agriculture, Saba Basha, Alexandria University, Egypt; [2]Central Laboratory for Aquaculture Research (CLAR), Agricultural Research Centre (ARC) and [3]Plant Production Department (Horticulture- Pomology), Faculty of Agriculture, Saba Basha, Alexandria University, Egypt.

DOI: 10.21608/jalexu.2021.111114.1028

Article Information

Received:December 10th 2021

Revised: December 12nd 2021

Accepted:December 27th 2021

Published: December 31st 2021

ABSTRACT: This research aimed to investigate the influence of Arbuscular Mycorrhizal Fungi (AMF) species *Rhizoglomus macrocarpium* (GM)*; Rhizoglomus irregularis* (GI) and *Rhizoglomus fasciculatum* (GF) inoculation, under alkaline soil conditions, on the Respiration of Microbial Soil (RMS) through the emission of CO_2, in an attempt to increase soil health and subsequently the plant growth and productivity. Nitrogen (N) and Potassium (K) fertilizers were applied at the rate of 30 g NH_4NO_3 and 20 g K_2SO_4 per seedling. Three levels of phosphorus fertilizer (calcium superphosphate (15.5 % P_2O_5)) were applied before filling the pots. In AMF pots, one week before planting, the soil was inoculated with 30 ml of AMF inoculums. Also, 20 ml of AMF inoculums were added with the seedlings of guava. Every day, tap water was used to irrigate all of the pots. The guava seedlings were harvested 120 days after planting. The results showed that all AMF species were effective on plant growth and P uptake in guava (*Psidium guajava L.*) seedlings. The AMF species GI developed the plant growth and P content of guava seedlings compared to other AMF species. The guava (*Psidium guajava L.*) seedlings inoculation with AMF led to increased soil microbial activity this is supported by evidence of increased RMS and CO_2, this resulted in improving soil health and increasing the availability of elements in the soil, resulting in increased P uptake by plants and increased plant growth.

Keywords: Guava; Mycorrhizal fungi; P levels; Soil microbial respiration

INTRODUCTION

Guava (*Psidium guajava L.*) is a main horticultural crop throughout the tropical and subtropical zone (Das et al., 2017). Guava is widespread fruit in Egypt, as it is inexpensive and contains high levels of vitamin C, pectin, vitamin A, B2, and minerals such as phosphorus (P), calcium (Ca), and iron (Fe) (Ibrahim et al., 2010). Furthermore, guavas can grow in newly reclaimed soils due to their high adaptability and thrive in these soils (Ibrahim et al., 2010).

The most common symbiosis between plants and fungi of the phylum *Glomeromycota* is arbuscular mycorrhizal fungi (AMF) (Karliński, 2021). Approximately 80% of plant species have a symbiosis relationship with AMF (Harley and Harley 1987). AMF increases the plant root surface and subsequently increases the absorption of P as they stimulate the mineralization of organic P (Barea et al., 2008).

Estrada-Luna et al., (2000) discovered that guava plants treated with a mixture of *Glomus diaphanum*, *Glomus albidum*, and *Glomus claroides* had greater plant growth parameters, shoot contents of P, Mg, Cu, and Mo, and an increase in gas exchange, than plants non-inoculated with AMF. Das et al., (2017) found that guava plants treated with *Glomus mosseae* increased guava plant yield. Similarly, Quiones-Aguilar et al., (2020) AMF studied in guava trees as growth stimulants, their results revealed a differential influence on guava plant growth when treated with native AMF, with colonization percentages of AMF exceeding 60%.

The ability of AMF species to improve plant growth varies (Tawaraya et al., 2012). Heidari and Karami, (2014) studied the effects of two AMF species (*G. mosseae*, and *G. etanicatum*) on sunflower yield, uptake of P, and oil content, under field circumstances and found that *G etanicatum* had the highest impact on seeds yield, P and oil content in seeds. Similarly, the impact of three species of AMF on the growth of specific vegetable crops grown in alkaline soils was investigated by Abou El Seoud et al., (2017).They found that the AMF species, such as *G. irregularis*, increased the plant growth and content of P in

squash and tomato, while *G. macrocarpium* enhanced the plant growth and content of P carrot plants under low P level.

The respiration of microorganisms in the soil (RMS) is one of the indexes of microbial activity in the soil (Biabani and Gholizadeh, 2020). It is one of the most important soil fertility indicators (Nannipieri et al., 2017). AMF increases RMS in temperate forests and agricultural soils by affecting soil CO_2 fluxes (Tomè et al., 2016). Also, Ardestani et al., (2019) found that RMS rates increased by using AMF.

In this context, the present work aimed to investigate the influence of AMF inoculation, under alkaline soil conditions, on the RMS through the emission of CO_2, to increase soil quality and subsequently the plant growth.

MATERIALS AND METHODS

Soil preparation

The soil sample was collected from the topsoil (0–30 cm) of Rosetta City, Bahira, Egypt. To homogenize and remove the roots and impurities from the soil sample, it was air-dried, crushed, and sieved through a two mm sieving size (No. 10 mesh) before being blended. The soil chemical properties were analyzed according to Page et al., (1982) as following: Soil texture is clay, pH (saturated soil paste extract) 7.81, EC (1:1) 0.82 dS/m, organic matter (OM) 1.72%, $CaCO_3$ 1.27%, available N 91 mg/kg soil, available P (Olsen) 7.8 mg/kg soil.

Plant materials

Guava seedlings were obtained from Al-Busaili Agricultural Research Station, Ministry of Agriculture and Soil Reclamation, Egypt. The seedlings were about three months old twenty centimeters high and had one branch.

Arbuscular Mycorrhizal Fungi (AMF) inoculation

The applied AMF species were *Rhizoglomus macrocarpium* (GM)*; Rhizoglomus irregularis* (GI) and *Rhizoglomus fasciculatum* (GF). The first and third species were gained from the department of plant nutrition, Göttingen-University- Germany. The second species was acquired from the department of plant pathology, Hanover University - Germany. These AMF species are activated in the Soil Microbiology lab - Soil and Agriculture Chemistry Dep, Faculty of Agriculture - Saba Basha, Alexandria University, Egypt.

Pot experiment

Plastic pots (20 cm in diameter and 18 cm depth) were washed, labeled, and a filter paper was placed on the bottom of all pots to prevent soil seep, then 3 kg soil was placed in each pot leaving an upper distance of about 5 cm without soil Then compact it until reaching a bulk density of 1.42 g cm^{-3}. One week before planting, all pots were irrigated to the volumetric moisture content of 0.25 cm^3cm^{-3}, which equals 70% of soil field capacity. Later, seedlings of guava were transplanted in pots on March 6, 2020. Nitrogen (N) and Potassium (K) fertilizers were applied at the rate of 30 g $NH4NO_3$ and 20 g K_2SO_4 per seedling. The N fertilizer was applied at four equal doses at the rate of 7.5 g/60 ml water for each pot. The K fertilizer was applied before putting the soil in the pots. After that, the phosphorus fertilizer (calcium superphosphate 15.5 % P_2O_5) were applied at three levels before filling the pots to obtain P0 (zero P fertilizer); P1 (half of the recommended P fertilizer (15 g / seedling) and P2 (all the recommended P fertilizer (30 g /seedling) (Hernandes et al., 2012). In AMF pots, one week before planting, the soil was inoculated with 30 ml of AMF inoculums (Malibari et al., 1990). Also, 20 ml of AMF inoculums were added with the seedlings of guava, (in total, the rate of 500 A-mycorrhizal spores per pot). Every day, tap water was used to irrigate all the pots to achieve 70% of field capacity.

Plants harvesting and analysis

The guava seedlings were harvested 120 days after planting (the harvest date July 7, 2020). At the harvest time, shoots of all guava seedlings were separated from roots. The guava shoots and half roots (by weight) were washed with tap water, distilled water, air dried, and over-dried at 70°C for 48 hours (Steyn, 1959) to constant weight, and the dry weight of the guava shoots and half roots were recorded, then ground in a mill and stored in a paper package for chemical analysis. Samples of plant material (shoots and half of the roots) were wet digested with H_2SO_4–H_2O_2 (Lowther, 1980). The vanadomolybdophosphoric method was used to determine the phosphorus concentration (Jackson, 1967).

Quantifying roots length (RL)

The remaining half of the plant roots were removed from the soil using a 0.5 mm screen and a stream of tap water. The roots were covered in layers of paper towels for 2-3 minutes to soak out any remaining moisture (Schenk and Barber, 1979). Three roots' samples weighing 0.3 g fresh weight were used to calculate the (RL) using line intersect method for each pot (Tennant, 1975).

$$RL(cm) = \frac{RFW}{0.1g} \times N \times 1.5714$$

Where RL= root length, RFW = root fresh weight (g), N =sum of horizontal and vertical crossing.

AMF extraction and determination

The root samples of guava seedlings were divided into 1 cm segments for each pot. About 100 root segments per pot were cleaned using 10% KOH at 90 °C for 15 min and acidified in 1% HCl for 10–15 min, then stained with (0.05%) trypan blue (Gemma et al., 1989). The AMF root colonization percentage was measured under the

microscope at 200X magnification (McGonigle et al., 1990). The AMF chlamydospores were extracted by using the electric wet sieving method described by Brundrett et al., (1996). 50 g of soil from each pot treated with AMF was suspended in a sufficient amount of water and stirred with rod glass for 1 min. The suspension was passing through the sieve set with sizes 500, 250, 100, 40 meshes, respectively. The AM spores in the last two sieves were transferred on the nylon filter with a 45 µm pore size. The enumeration of AM spores was done for each sample three times under a light microscope with 40X magnification. The number of AMF spores / g soil was recorded.

Respiration of microbial soil (RMS)

At 25°C, soil samples are incubated in a closed vessel. The CO_2 produced is absorbed in sodium hydroxide and titrated to determine its concentration (Isermeyer, 1952, and modified by Jaggi, 1976). Weigh 20 g of field-moist soil into three centrifuge tubes or test tubes (samples). Pipette 20ml of sodium hydroxide solution into the laboratory bottles and insert the tubes into the bottles. Close the bottles and incubate for 24 h at 25°C .Remove the tubes and add 2ml of barium chloride solution to precipitate the absorbed CO_2 as barium carbonate. Add 3-4 drops of indicator solution and titrate the remaining sodium hydroxide with dilute HCl .To prepare controls, perform the procedure without soil.

Available Phosphorus soil

Available P was extracted from the soil at the harvest time using the $NaHCO_3$ (0.5 N, pH = 8.5) method (Olsen et al., 1954) and measured by ascorbic acid molybdenum blue method at Spectrophotometer wavelength 406 nm (Murphy and Riley 1962).

Statistical methods:

The treatments as one factor were arranged in a randomized complete block design (RCBD) with six replicates, according to Gomez and Gomez (1984). The treatments were compared using Duncan's LSD at a 5% level of probability using the CoStat computer program (CoStat, Ver. 6.311, 2005).

RESULTS

1. Plants dry weight

At all P levels, there was no significant difference in shoot dry weight of guava seedlings treated with AMF species (GI).

At all P levels, the dry weight of all plants treated with the three species of AMF improved significantly as compared to plants not inoculated with AMF (Fig 1). In other words, the response of the plants' dry weight without AMF inoculation to increase P levels was higher than the other plants treated with AMF species. The plants' dry weight of guava seedlings without AMF at a high P level, increased by (1.64-fold) compared to the guava seedlings at the first P level (P0). Whereas the plants dry weight of guava seedlings inoculated with species of AMF GM, GF, and GI at the third P level (P2), increased by only (1.34-fold; 1.35-fold; and 1.1-fold) compared to the guava seedlings inoculated with the same AMF species respectively at the first P level.

The greatest plant dry weight of guava seedlings was observed with (GI) species, while inoculation with the AMF species (GM) and (GF) led to higher plant dry weight than that of the control (uninoculated plants) but significantly lower than the other guava seedlings inoculated with (GI) specie. The plants' dry weight of guava seedlings inoculated with AMF species (GI) increased by (3.18-fold; 1.76-fold; and 1.82-fold) compared to the guava seedlings without AMF inoculation, with AMF species (GM), and (GF) respectively at the first P level and increased by (2.14-fold; 1.45-fold; and 1.49-fold) compared to the guava seedlings without AMF inoculation, with AMF species (GM), and (GF) respectively at the third P level.

At each P level, there was no significant difference in plant dry weight of guava seedlings between AMF species (GM) and (GF).

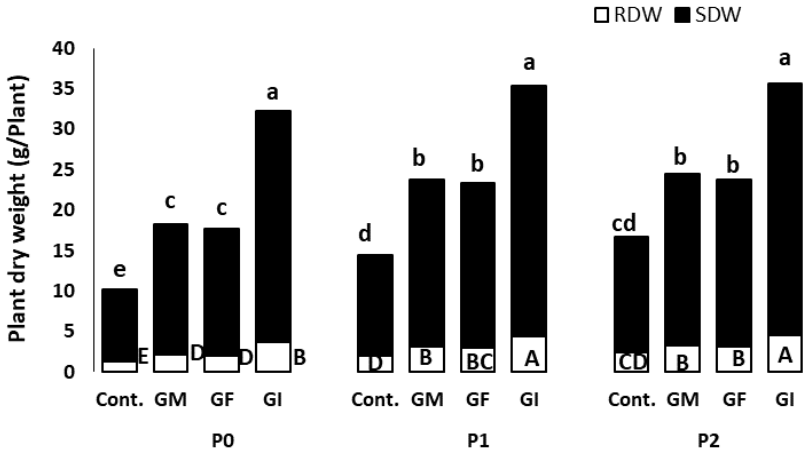

Fig. (1): Plant dry weight (g/ plant) of guava seedlings as affected by P levels and inoculated with AMF species (GM)*;* (GF), and (GI); different letters indicate a significant difference between AMF species at all P levels, small letters between shoot dry weight and capital letters between root dry weight, P≤0,05).

Root length (RL)

As shown in (Fig 2), at the high P level, the root length (RL) of guava seedlings with and without AMF inoculation grew significantly. The RL of guava seedlings that were not inoculated with AMF had a stronger response to increasing P levels than the other plants treated with AMF species. The RL of guava seedlings without AMF species at a high P level increased by (1.94-fold) compared to the guava seedlings without AMF at the first P level (P0), whereas the RL of guava seedlings inoculated with AMF species at the third P level, increased by (1.46-fold; 1.57-fold; and 1.51-fold) only as compared to RL of guava seedlings treated with the same AMF species respectively at a low P level.

The guava seedlings inoculated with (GI) produced the longest RL compared to the other plants treated with (GM) and (GF) at all P levels. When its values increased by (3.4-fold; 1.75-fold; and 1.9-fold) compared to RL of guava seedlings without AMF inoculation, with AMF species (GM), and (GF) respectively at low P level and increased by (2.5- fold; 1.7-fold; and 1.76-fold) compared to RL of guava seedlings without AMF inoculation, with AMF species (GM), and (GF) respectively at the third P level.

When comparing the RL of guava seedlings treated with AMF species (GM) to the RL of guava seedlings treated with AMF species (GM) at each P level, there was no significant difference.

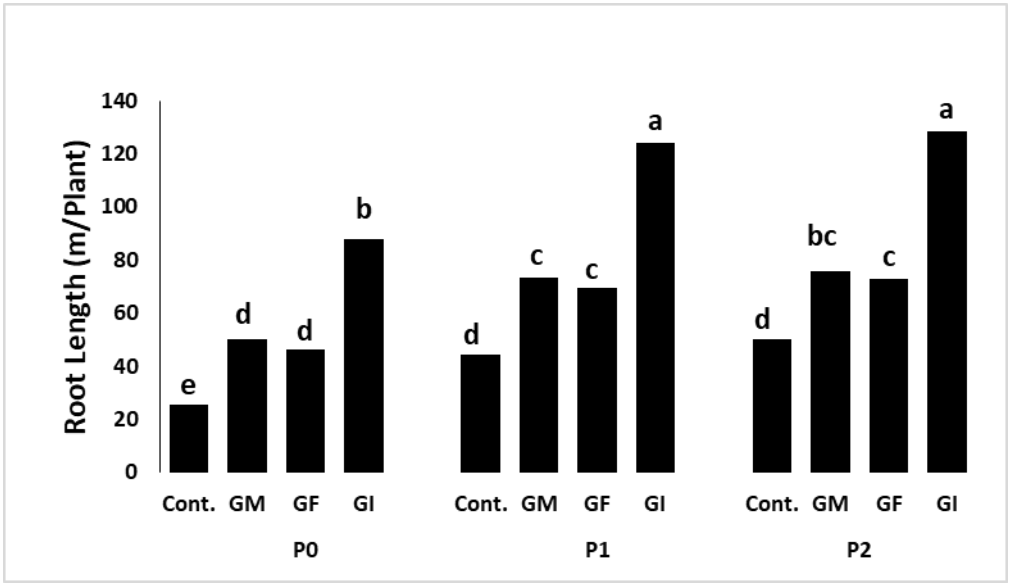

Fig. (2): Root length (m/ plant) of guava seedlings as affected by P levels and inoculated with AMF species (GM)*;* (GI), and (GF); different letters indicate significant differences between AMF species at all P levels, P≤0,05).

3. Respiration of microbial soil (RMS)

At all P levels, RMS treated with AMF species was a highly significant increase compared to non-treated soils. RMS of plants treated with AMF species (GM)*;* (GF), and (GI) increased by (25.8-fold; 25.4-fold; and 34.3-fold) respectively compared to non-treated soil at the first P level (Fig 3).

There was no significant difference in RMS of AMF species between the two high P levels.

RMS was significantly higher in AMF species (GI) compared to the AMF species (GM) and (GF) at all P levels. RMS with AMF specie (GI) increased by (1.32-fold; and 1.35-fold) compared to (GM)*;* (GF) respectively at low P level and it was increased by (1.33-fold; and 1.4-fold) compared to (GM)*;* (GF) respectively at high P level (Fig 3).

There was no significant difference in RMS between plants treated with AMF species (GM) and other plants treated with AMF species (GF) at each P level (Fig 3).

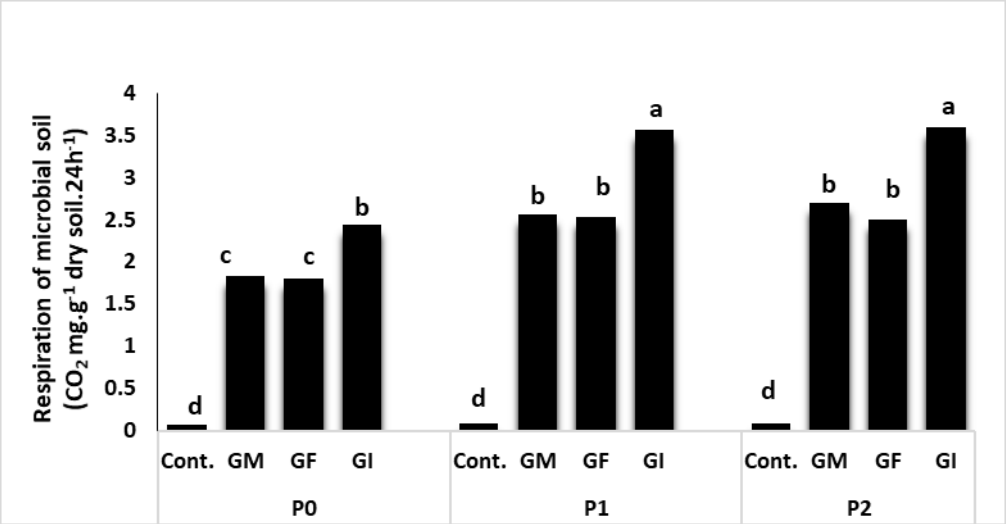

Fig. (3): RMS (CO_2 mg. g^{-1} dry soil.$24h^{-1}$) as affected by P levels and inoculated with AMF species (GM)*;* (GI) and (GF); different letters indicate significant differences between AMF species at all P levels, P≤0,05).

4. AMF measurements

4.1.AMF root colonization % (RCP)

At the third P level, the RCP of all AMF species of guava seedlings lowered significantly (Fig 4). In other words, at low P levels, the RCP of AMF species (GI), (GM), and (GF) increased by around (1.47-fold; 1.44-fold; and 1.45-fold) respectively, compared to the same AMF species at high P levels.

The mycorrhizal root colonization % was significantly higher in AMF species (GI) compared to other AMF species (GM and GF) at all P levels. On the other hand, there was no significant difference at each P level between RCP of (GM and GF) species at all P levels.

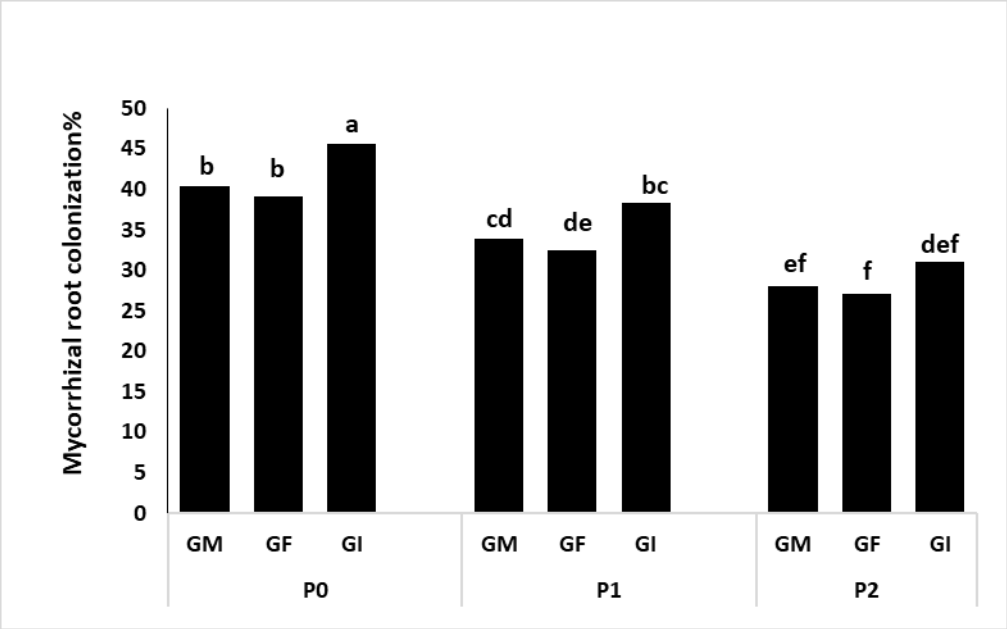

Fig. (4): Mycorrhizal root colonization % as affected by P levels and inoculated with AMF species (GM)*;* (GI), and (GF); different letters indicate significant differences between AMF species at all P levels, P≤0,05).

4.2. Number of AMF spores

Significant difference was observed in the number of AMF spores as a result of increasing P levels at all AMF species (Fig 5). The number of AMF spores at GM, GF, and GI species at the lowest P level (P0) reduced by about (151%; 153%; and 143%) respectively as compared to the highest P level (P3).

At all P levels, the amount of AMF spores in guava plant roots of the AMF species (GI) was significantly higher than the other two AMF species (GM and GF). On other hand, at all P levels, there was not a significant difference in AMF spores' number between the AMF species (GM and GF) (Fig 5).

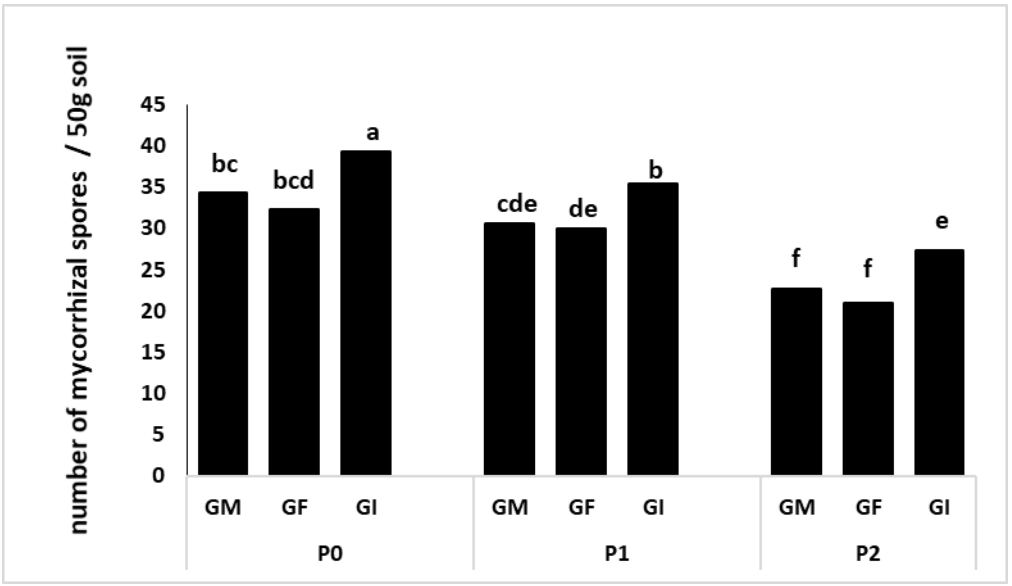

Fig. (5): Number of AMF spores/50 g soil as affected by P levels and inoculated with AMF species (GM)*;* (GI), and (GF); different letters indicate significant differences between AMF species at all P levels, P≤0,05).

Available phosphorus in both plant and soil

5.1. Plant phosphorus content

The results clearly showed that the plant P concentration of guava seedlings inoculated and uninoculated with AMF improved with increasing P levels (Fig 6). Uninoculated guava seedlings had a stronger response to increasing P levels than the other plants inoculated with the other AMF species studied. The plant P concentration of guava seedlings uninoculated and inoculated with AMF species (GI); (GM) and (GF) at the third P level increased by (1.7-fold; 1.23-fold; 1.27-fold; and 1.19- fold) compared to uninoculated and inoculated with AMF species (GI); (GM) and (GF) respectively at the first P level (Fig 6). On the other hand, plant P concentrations of guava seedlings inoculated with all AMF species at the second P level compared to the third P level did not differ significantly.

Among the AMF species studied, (GI) led to a highly significant in the plant P concentration of guava seedlings as compared to the other two AMF species (GM and GF) at all P levels (Fig 6). There was no significant difference in plant P content of guava seedlings between AMF species (GM and GF) at each P level (Fig 6).

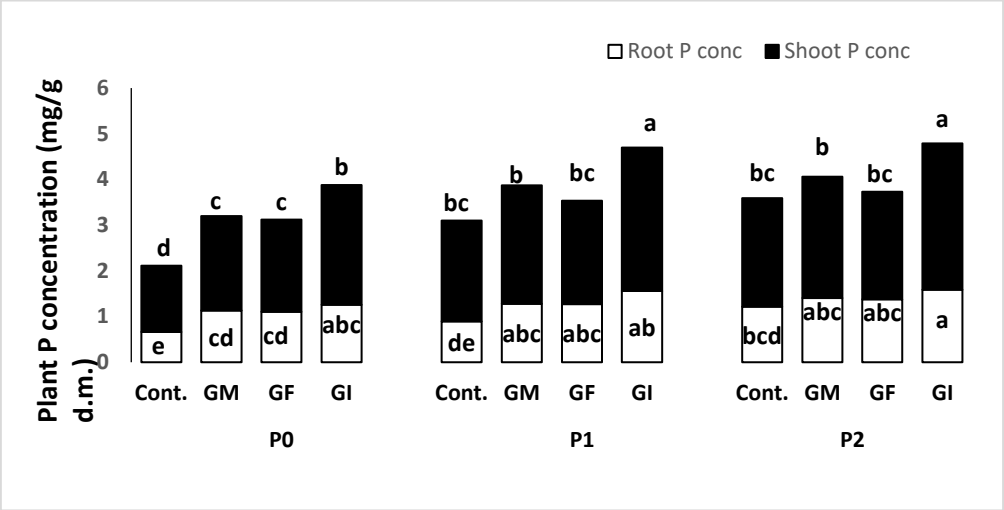

Fig. (6): Shoot and root P concentration (mg P/ g.d.m.) of guava seedlings as affected by P levels and inoculated with AMF species (GM)*;* (GI) and (GF); different letters indicate a significant difference between AMF species at all P levels, small letters between shoot P concentration and capital letters between root P concentration, P≤0,05).

5.2. Plant phosphorus uptake

The plant P uptake of guava seedlings inoculated and uninoculated with AMF increased with increasing P levels (Fig 7). Also, the response of uninoculated guava seedlings to increase P levels was greater than that of the other plants treated with all studied AMF species. The plant P uptake of guava seedlings uninoculated and inoculated with all AMF species at the third P level increased by (2.65-fold; 1.34-fold; 1.72-fold; and 1.56-fold) compared with uninoculated and inoculated with AMF species (GI); (GM), and (GF) respectively at the first P level (Fig 7). On the other side, there was not a significant difference in P uptake of all AMF species treated guava seedlings at the second P level compared to the third P level.

At all P levels, the AMF species (GI) led to a highly significant in the plant P uptake of guava seedlings as compared to the other two AMF species (GM and GF) (Fig 7). Also, there was not a significant difference in plant P uptake of guava seedlings treated with AMF species (GM) as compared to plant P uptake of guava seedlings treated with AMF species (GF) at all P levels.

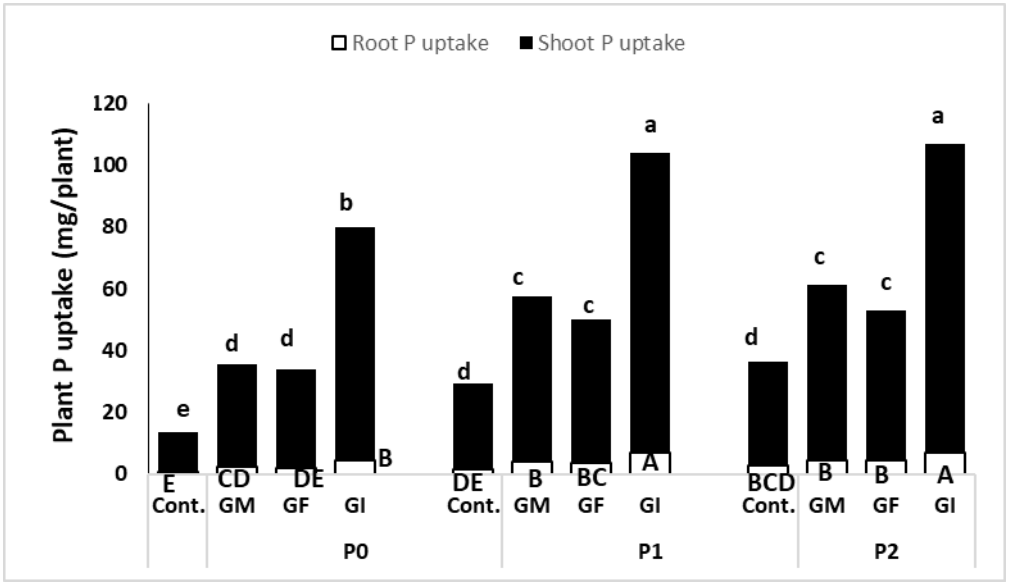

Fig. (7): Shoot and root P uptake (mg P/ plant) of guava seedlings as affected by P levels and inoculated with AMF species (GM)*;* (GI), and (GF); different letters indicate a significant difference between AMF species at all P levels, small letters between shoot P uptake and capital letters between root P uptake, P≤0,05).

5.3. Available phosphorus in soil

With rising P levels, the available P in the soil of plants treated and untreated with AMF was significantly higher. (Fig 8). The available P in soil inoculated and uninoculated with AMF increased with increasing P levels. The soil available P of plants uninoculated and inoculated with AMF species (GI),(GM) and (GF) at the third P level increased by (2.35-fold; 1.38-fold; 1.48-fold; and 1.6-fold) as compared with uninoculated and inoculated with AMF species (GI); (GM) and (GF) respectively at the first P level (Fig 8).

At the first P level, inoculated guava plants with different AMF species resulted in a highly significant increase in soil available phosphorus but responded differently to the different species of AMF. The available P in the soil of plants inoculated with AMF species (GI); (GM) and (GF) at the first P level increased by (1.85-fold; 1.5-fold; and 1.34-fold) respectively as compared to without AMF inoculation (control) at the first P level (P0). On the other side, at the third P level, there was no significant variation in soil available P of plants treated with AMF species as compared to plants without AMF inoculation.

Among the AMF species studied, (GI) led to a highly significant increase in available soil phosphorus as compared to the other AMF species at all P levels (Fig 8). At all P levels, there was no significant difference was observed between AMF species (GM) and (GF) in soil available phosphorus. There was a significant high in soil available phosphorus of plants treated with AMF species (GI, GM, and GF) at the first P level as compared to the third P level.

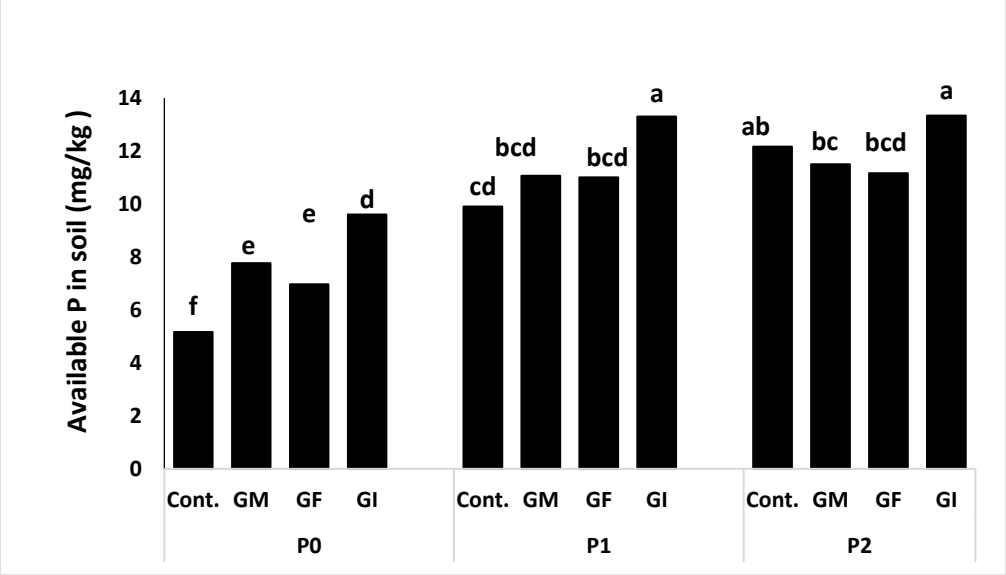

Fig. (8): Available P in soil (mg P/ kg soil) as affected by P levels and inoculated with AMF species (GM)*;* (GI) and (GF); different letters indicate significant differences between AMF species at all P levels, P≤0,05).

DISCUSSIONS

The plants' growth parameters such as shoot dry weight, root dry weight, and (RL) treated with AMF increased significantly in comparison to other guava seedlings with no AMF inoculation at all P levels. The supply of P fertilizer to the soil led to significantly enhanced uninoculated plant growth parameters. However, when the soil was treated with AMF, the effect of P fertilizer on plants' growth parameters remained less pronounced. This result is consistent with Oseni et al., (2010), and Abou El Seoud et al., 2017. The same results were observed with guava (Ratna and Bahadur, 2019). Sato et al., (2019) found that the effect of P fertilizer on plants' growth parameters treated with AMF remained less pronounced compared to the other plants without AMF inoculation. This may be because AMF colonization was reduced in soils with high soluble phosphate concentrations (Kaeppler et al., 2000). Increased P availability in the soil resulted in decreased AMF colonization, spore generation, and AMF hyphae length (Valentine et al., 2001). That may be due to improve levels of phospholipids ,which reduce membrane permeability and decrease exudation of amino acids, organic acids, and sugars, which are the major source of nutrition for the growth and development of germinating AMF spores (Ratnayake et al., 1978). It can be concluded, AMF has a limited impact when the P level has high (Abou El Seoud et al., 2020). In the same line, Neumann and George (2005) found that the plants treated by the AMF root system of plants were developed when the P level was low. AMF may also enhance the root surface area (SA) of plants (Aguim et al., 2004), which leads to enhanced uptake of nutrients. Plants with large root systems can absorb more nutrients and water from the soil and grow faster compared to plants with small root systems (Abou El Seoud, 2008). In contrast, when comparing plants inoculated with AMF to other uninoculated plants, Bonanomi et al., (2001) found that there were no significant variations at the root system.

The greatest plants growth parameters effects in guava seedlings were observed with (GI), and insemination with two AMF species (GM) and (GF) led to greater plant growth parameters compared to uninoculated plants but significantly lower than the other guava seedlings inoculated with (GI) specie. Similarly, Drew et al., (2003) found that plants inoculated by G. *intraradiaces* grew better than plants treated by G. *mosseae*. In a similar vein, Amer et al. (2010) found that bean plants treated with the AMF species G. *intraradiaces* grew significantly increased than those treated with the other AMF species Glomus *macrocarpium.* The effort of AMF to develop the growth and quality of their host plants is influenced by the host species, the AMF species, and their interactions, according to Baum et al., (2015). These results are consistent with those of Kim et al., (2017). Previous research indicated that plant development differed according to the species of AMF inoculant used (Abou El Seoud et al., 2017).

In the present study, there were highly significant increases in RMS treated with AMF species compared with non-treated soils at all P levels. The AMF provides the plants with nutrients and water (Smith and Read, 2008). On the other hand, plants provide carbon (C) to AMF as well as another microorganisms in the root zone. So, AMF increased the respiration of microbial soil (Lang et

al., 2020). Tomè et al., (2016) found that AMF can significantly affect the respiration of microbial soil. These results are consistent with Zhang et al., (2016). Also, Lang et al., (2020) found that AMF improved the respiration of microbial soil. On the contrary, Raiesi and Ghollarata (2006) found that decreases respiration of microbial soil in plants treated with AMF.

The present study found that all measured AMF parameters (RCP and number of AMF spores) of treated with AMF species decreased significantly with increasing P levels. Kahiluoto et al., (2001) observed reduces in AMF colonization densities with increasing P levels. Also, Ryan and Graham (2002) reported that a high amount of available P in soil limits AMF colonization and inhibits spore germination. This result agrees with Prasad et al., (2012), Carine et al., (2017), Sato et al., (2019), and (Ziane et al., 2021). This could be attributed to a decrease in the excretion of compounds responsible for AMF hyphal growing and the colonization of AMF due to the high phosphorus content in plant root tissue (Nagahashi and Douds, 2000). Moreover, cell phospholipids affecting on membrane permeability and emission of carbohydrate compounds that are necessary for AMF association (Schwab et al., 1991), which leads to reduces in AMF colonization densities and inhibits spore germination (Ryan and Graham, 2002).

This study found that the plant P content and plant P uptake of guava seedlings inoculated and uninoculated by AMF increased with increasing P levels. The response of uninoculated guava seedlings to increase P level was stronger than that of the other plants treated by several AMF species. On other hand, the plant P concentration and uptake were improved by AMF inoculation especially at low P level compared to plants without AMF. These results are consistent with Gupta et al., (2002), inoculating plants with AMF enhanced their P content and uptake, which could be related to depletion of accessible P surrounding the soil rhizosphere as compared to uninoculated plants. In addition, Artursson et al., (2006) found that plants treated with AMF accumulated considerably more phosphorus (P) in plant tissues than control plants (uninoculated plants). In the same direction, Meddad-Hamza et al., (2010) discovered that plants inoculated by AMF exhibited higher P content than untreated plants. Similarly, Dennett et al., (2011) found that treated plants with AMF significantly improved root phosphorus content. These results agree with Püschel et al., (2021). That could be because AMF increases the root surface area (SA) of the plants by improving the first-degree lateral root branches (Aguim et al., 2004), which leads to increased nutrient uptake by extending the soil depletion zone around the root system (Aguim et al., 2004). A major effect of the AMF on plants growth is the reduction of a P shortage (Tawaraya et al., 2012 and Liu et al., 2014). Another hypothesis ascribed the increased P-content to the AMF plants' greater total root system or efficiency, which would undoubtedly lead to increased total nutrient intake (Abou El Seoud et al., 2020).

Among the AMF species studied, (GI) led to a very significant rise in plant P content and plant P uptake of guava seedlings as compared to the other two AMF species (GM and GF) at all P levels. Heidari and Karami (2014) investigated the impacts of two different mycorrhizae species, *G. mosseae*, and *G. etanicatum*, on grain yield, uptake of nutrients, and oil content in sunflower plants, and found that *G. etanicatum* was the greatest impact on grain yield and nutrient content in plants. In addition, Abou-El-Seoud et al. (2017) found that squash and tomato plants treated with AMF species *G. intraradiaces* were higher plant P uptake compared to those inoculated by other species of AMF. In contrast, Akay et al., (2016) found that the effects of several species of AMF (*G. geosporum*, *G. mosseae*, *G. caledonium*, and *G. etunicatium*) on the plant nutrients content of lupin (*Lupinus albus L.*) and found no significant differences between the different species of A-mycorrhizal fungi treated to the lupin plant.

The present study found that the plants without AMF had a highly significant increase in available P in soil with increasing P levels. Similarly, Abou-El-Seoud et al., (2018) discovered that the available phosphorus in the soil of plants not inoculated by AMF had a strong response to an increase in P levels compared to plants treated with AMF. At a high P level, there was no significant response at different AMF species in soil available phosphorus compared with untreated plants (control). On the contrary, when the P level was low, plants treated by different AMF species were a very significant increase in soil available phosphorus, but it was responding differently to the different AMF species. These findings are consistent with those of Abou-El-Seoud et al., (2017). That could be because the AMF's hyphae create organic acids and Phosphatase, which work on the availability of phosphorus from organic complexes, improving soil P availability (Aono et al., 2004). AMF hyphae also cause a drop in pH of the alkaline soil from 8.5 to 7.4 in alkaline soils by exuding organic acids, which work to make nutrients available, such as phosphorous (Giri et al.,2005). The hyphae of AMF stretch into the soil away from the root surface or root hair area (up to 25 cm). Their small diameter (equal or less than root hairs) helps the AMF to stretch into soil pores that are inaccessible to roots with much larger diameters. This indicates they can use solution-filled soil pores at much lower soil water potentials

than roots, allowing them to uptake phosphorus from drier soils (Jakobsen et al., 2005).

In conclusion, based on the findings of this research and under the same working conditions, we may recommend that all AMF species were effective on plant growth and P uptake in guava (*Psidium guajava L.*) seedlings. The AMF species GI developed the plant growth and P content of guava seedlings compared to other AMF species. The guava (*Psidium guajava* L.) seedlings inoculation with AMF led to increased soil microbial activity this is supported by evidence of increased RMS and CO_2, this resulted in improved soil health and increased the availability of elements in the soil, resulting in increased P uptake by plants and increased plant growth.

REFERENCES

Abou El Seoud, I I (2008). Phosphorus efficiency of tagetes plant inoculated with two arbuscular mycorrhizal fungi strains. Aust. J. Basic and App. Sci., 2(2): 234- 242.

Abou El Seoud, I I; M M Yousry and N M Abd El Hamid (2017). Effectiveness of three arbuscular mycorrhizae species on growth of some vegetable crops under calcareous soil conditions. *Egyptian Academic Journal of Biological Sciences, H. Botany*, *8*(1), 33-47.

Abou El Seoud, I I; H A Mahmoud and R M ELadly (2018). Phosphorus efficiency of wheat (Triticum aestivum) genotypes inoculated with mycorrhizal fungi under calcareous soil conditions. *Egyptian Academic Journal of Biological Sciences, G. Microbiology*, *10*(1), 21-35.

Abou El Seoud, I I; N M Abd El Hamid and S W Mahmoud (2020). Mycorrhizae can support squash plant growth in Phosphorus deficient calcareous soil. *Egyptian Journal of Soil Science*, *60*(4), 425-435.

Aguim, O; J P Mansilla; A Vilarino and M J Sainz (2004). Effects of mycorrhizal inoculation on root morphology and nursery production of three grapevine rootstocks. American J Enology and Viticulture 55: 108-111.

Akay, A; M Yorgancilar and E Atalay (2016). Effects of different types of mycorrhiza on the development and the elemental content of lupin (Lupinus albus L.). *Journal of Elementology*, *21*(2).

Amer, M A; I I Abou El Seoud; M R Rasmy and Manar M Khater (2010). Effects of A-Mycorrhizal fungi, bacteria, *and yeast as a biological control of sclerotinia* sclerotiorum, on the growth of common bean (Phaseolus vulgaris L.). Alex. Sci. Exc. J. 31(4): 339-351.

Aono, T; I E Maldonado-Mendoza; G R Dewbre; M J Harrison and M Saito (2004). Expression of alkaline phosphatase genes in arbuscular mycorrhizas. *New Phytologist* .162: 525–534.

Ardestani, M M; V Jílková; M Bonkowski and J Frouz (2019). The effect of arbuscular mycorrhizal fungi Rhizophagus intraradices and soil microbial community on a model plant community in a post-mining soil. Plant Ecology, 220(9), 789-800.

Artursson, V; R D Finlay and J K Jansson (2006). Interactions between arbuscular mycorrhizal fungi and bacteria and their potential for stimulating plant growth. Environ. Microbiol. 8: 1–10.

Barea, J M; N Ferrol; C Azcon –Agular and R Azcon (2008). mycorrhizal symbioses In: white pl Hammond J P (eds)The Ecophysiology interactions Vol 7plant Ecophysiology Seris Springer Drodrecht,pp 143-163.

Baum, C; W El-tohamy and N Gruda (2015). Increasing th productivity and productu quality of vegetable crops using arbuscular mycorrhizal fungi. Scientia Horticultuae187: 131-141.

Biabani, A and A Gholizadeh (2020). Study of microbial respiration in different types of vermicompost. Malaysian Journal of Soil Science, 24, 135-146.

Bonanomi, A; J H Oetiker; R Guggenheim; T Boller; A Wiemken and R Vögeli-Lange (2001). Arbuscular mycorrhiza in mini-mycorrhizotrons: first contact of Medicago truncatula roots with Glomus intraradices induces chalcone synthase. *New phytologist*, *150*(3), 573-582.

Brundrett, M; N Bougher; B Dell; T Grove and N Malajczuk (1996). *Working with mycorrhizas in forestry and agriculture* (Vol. 32, p. 374). Canberra: Australian Centre for International Agricultural Research.

Carine, T N; W Germaine-Alice; T V Desire; M T Judith; O A Neree; Y Emmanuel and N N Godswill (2017). Effect of phosphorus fertilization on arbuscular mycorrhizal fungi in the Bambara groundnut rhizosphere. *African Journal of Microbiology Research*, *11*(37), 1399-1410.

Das, K; S Sau; P Datta and D Sengupta (2017). Influence of bio-fertilizer on guava (Psidium guajava L.) cultivation in gangetic alluvial plain of West Bengal, India. *Journal of Experimental Biology and Agricultural Sciences*, *5*(4), 476-482.

Dennett, A L; LW Burgess and M H Ryder (2011). Arbuscular mycorrhizal associations in salanum centrale (bush tomato operennial sub-shrab from the arid zone of Australia. journal of Arid Environments75: 688-694.

Drew, E A; R S Murray; S E Smith and I Jakobsen (2003). Beyond the rhizosphere: growth and function of arbuscular mycorrhizal external hyphae in sands of varying pore sizes. 251: 105-114.

Estrada-Luna, A A; Jr F T Davies and J N Egilla (2000). Mycorrhizal fungi enhancement of

growth and gas exchange of micropropagated guava plantlets (Psidium guajava L.) during ex vitro acclimatization and plant establishment. Mycorrhiza, 10(1), 1-8.

Gemma, J N; R E Koske and M Carreiro (1989). Seasonal dynamics of selected species of VA mycorrhizal fungi in a sand dune. *Mycological Research*, *92*(3), 317-321.

Giri, B; R Kapoor and K G Mukerji (2005). Effect of the arbuscular mycorrhizae Glomus fasciculatum and G. macrocarpum on the growth and nutrient content of Cassia siamea in a semi-arid Indian wasteland soil. New Forests, 29(1), 63-73 .

Gomez, K A and A A Gomez (1984). *Statistical procedures for agricultural research*. John Wiley & Sons.

Gupta, C; R Dubey and D Maheshwari (2002). Plant growth enhancement and suppression of Macrophomina phaseolina causing charcoal rot of peanut by fluorescent Pseudomonas Biol. Fertil. Soils. 35: 399-405.

Harley, J L and E L Harley (1987). A cheklist of the status mycorrhizal of the British flora. New Phytologist (Suppl.) 105.

Heidari, M and V Karami (2014). Effects of different mycorrhiza species on grain yield, nutrient uptake and oil content of sunflower under water stress. *Journal of the Saudi society of agricultural sciences*, *13*(1), 9-13.

Hernandes, A; S É Parent; W Natale and L É Parent (2012). Balancing guava nutrition with liming and fertilization. *Revista Brasileira de Fruticultura*, *34*(4), 1224-1234.

Ibrahim, H I M; M M A Zaglol and A M M Hammad (2010). Response of Balady guava trees cultivated in sandy calcareous soil to biofertilization with phosphate dissolving bacteria and/or VAM fungi. *J. Am. Sci*, *6*(9), 399-404.

Isermeyer, H (1952). Eine einfache Methode zur Bestimmung der Bodenatmung und der Karbonate im Boden. *Zeitschrift für Pflanzenernährung, Düngung, Bodenkunde*, *56*(1-3), 26-38.

Jackson, M L (1967). Soil chemical analysis. Prentic Hall, Inc., Engle wood cliff., USA.

Jaggi, W (1976). Die Bestimmung der CO_2-Bildung als Maβ der bodenbiologischen Aktivitat. *Schweiz Landwietschaft Forschung Band*, *15*(314), 317-380.

Jakobsen, I; B Chen; L Munkvold; T Lundsgaard; and Y G ZHU (2005). Contrasting phosphate acquisition of mycorrhizal fungi with that of root hairs using the root hairless barley mutant. *Plant, Cell and Environment*, *28*(7), 928-938.

Kaeppler, S M; J L Parke; S M Mueller; L Senior; C Stuber and W F Tracy (2000). Variation among maize inbred lines and detection of quantitative trait loci for growth at low phosphorus and response inverness to arbuscular mycorrhizal fungi. Crop Sci., 40: 358–364.

Kahiluoto, H; E Ketoja; M Vestberg and I Saarela (2001). Promotion of AM utilization through reduced P fertilization 2. Field studies. Plant Soil ,231, 65-79.

Karliński, L (2021). The Arbuscular Mycorrhizal Symbiosis of Trees: Structure, Function, and Regulating Factors. In *Symbiotic Soil Microorganisms* (pp. 117-128). Springer, Cham.

Kim, S J; J K Eo; E H Lee; H Park and A H Eom (2017). Effects of arbuscular mycorrhizal fungi and soil conditions on crop plant growth. *Mycobiology*, *45*(1), 20-24.

Lang, A K; F V Jevon; M P Ayres and J H Matthes (2020). Higher soil respiration rate beneath arbuscular mycorrhizal trees in a northern hardwood forest is driven by associated soil properties. *Ecosystems*, *23*(6), 1243-1253.

Liu, L; Z Gong;Y Zhang and P Li(2014). Growth, cadmium uptake and accumulation of maize (*Zea mays* L.) under the effects of arbuscular mycorrhizal fungi. Ecotoxicology 23: 1979-1986.

Lowther, G R (1980). Using of a single H_2So_4 - H_2O_2 digest for the analysis of Pinus radiate needles. Commun.Soil Sci. l.Analysis, 11: 175-188.

Malibari, A A; F A AL-Fassi and E M Ramadan (1990). Studies on VA-mycorrhizal of the western region soil, Saudi Arabia Annals Agric. Sci., Fac. Agric., Ain-Shams Univ., Cairo, Egypt. 35(1): 95-111.

McGonigle, T P; M H Miller; D G Evans; G L Fairchild and J A Swan (1990). A new method which gives an objective measure of colonization of roots by vesicular—arbuscular mycorrhizal fungi. *New phytologist*, *115*(3), 495-501.

Meddad-Hamza, A; A Beddiar; A Gollotte; M C Lemoine; C Kuszala and S Gianinazzi (2010). Arbuscular mycorrhizal fungi improve the growth of olive trees and their resistance to transplantation stress. Afr. J. Biotechnology. 9(8): 1159-1167.

Murphy,J and J P Riley (1962). Amodified single solution for the determipation of phosphate in natural waters, Anal. Chem. Acta. 27: 31-36.

Nagahashi, G and D D Douds (2000). Partial separation of root exudates components and their effects upon the growth of germinated spores of AM fungi. Mycol. Res., 104: 1453–1464.

Nannipieri, P; S greco and B Ceccanti (2017). Ecological significance of the biological activity in soil. *Soil biochemistry*, 293-356.

Neumann, E and E George (2005). Does the presence of arbuscular mycorrhizal fungi influence growth and nutrient uptake of a wild-type tomato cultivar and a mycorrhiza-defective mutant, cultivated with roots sharing the same soil volume? New Phytol., 166: 601-609.

Olsen, S C; F C Watanabe and L A Dean (1954). Estimation of available phosphorus in soils by

extraction with sodium bicarbonate. U. S. D.A Circular No.939.

Oseni, T O; N S Shongwe and M T Masarirambi (2010). Effect of arbuscular mycorrhiza (am) inoculation on the performance of tomato nursery seedlings in vermiculite. Int. J. Agric. Biol., 12:789–792.

Page, A L; D R Keeney; D E Baker; R H Miller; J Rosecoe Ellis and J D Rhoades (1982). Methods of Soil Analysis. American, Inc., Madison, Wisconsin, U.S.A

Prasad, K; A Aggarwal; K Yadav and A Tanwar (2012). Impact of different levels of superphosphate using arbuscular mycorrhizal fungi and Pseudomonasfluorescens on Chrysanthemum indicum L. *Journal of soil science and plant nutrition*, *12*(3), 451-462.

Püschel, D; M Bitterlich; J Rydlová and J Jansa (2021). Drought accentuates the role of mycorrhiza in phosphorus uptake. *Soil Biology and Biochemistry*, *157*, 108243.

Quiñones-Aguilar, E E; G Rincón-Enríquez and L López-Pérez (2020). Native mycorrhizal fungi as growth promoters in guava plants (Psidium guajava L.). Terra Latinoamericana, 38(3), 541-554.

Raiesi, F and M Ghollarata (2006). Interactions between phosphorus availability and an AM fungus (Glomus intraradices) and their effects on soil microbial respiration, biomass and enzyme activities in a calcareous soil. *Pedobiologia*, *50*(5), 413-425.

Ratna, S M and V Bahadur (2019). Effect of chemical fertilizers, bio-fertilizers and organic manure on growth, yield and quality of guava under Prayagraj agro-climatic condition. *Journal of Pharmacognosy and Phytochemistry*, *8*(4), 3154-3158.

Ratnayake, M; R T Leonard and J A Menge (1978). Root exudation in relation to supply of phosphorus and its possible relevance to mycorrhizal formation. *New Phytologist*, *81*(3), 543-552.

Ryan, M H and J H Graham (2002). Is there a role for arbuscular mycorrhizal fungi in production agriculture? Plant Soil, 244, 263–271.

Sato, T; S Hachiya; N Inamura; T Ezawa; W Cheng and K Tawaraya (2019). Secretion of acid phosphatase from extraradical hyphae of the arbuscular mycorrhizal fungus Rhizophagus clarus is regulated in response to phosphate availability. *Mycorrhiza*, *29*(6), 599-605.

Schenk, M K and S A Barber (1979). Phosphate uptake by corn as affected by soil characteristics and root morphology. Soil Science Society of American Journal 43: 880-883.

Schwab, S M; J A Menge and P B Tinker (1991). Regulation of nutrient transfer between host and fungus in vesicular mycorrhizas. New Phytol 117:387–398.

Smith, S E and D J Read (2008). Mycorrhizal symbiosis, 3rd edn. Academic.

Steyn, W J A (1959). Leaf analysis, errors involved in the preparative phase. J. Agric. Food Chem. 7: 344-348.

Tawaraya, K; R Hirose and T Wagatsuma (2012). Inoculation of arbuscular mycorrhizal fungi can substantially reduce phosphate fertilizer application to *Allium fistulosum* L. and achieve marketable yield under field condition. Biol. Fertil. Soils. 48: 839-843.

Tennant, D (1975). A test of a modified line intersect method of estimating root length. J. Ecol. 63: 995-1001.

Tomè, E; M Ventura; S Folegot; D Zanotelli; L Montagnani; T Mimmo and F Scandellari (2016). Mycorrhizal contribution to soil 1852 YUE ET AL. respiration in an apple orchard. Applied Soil Ecology, 101, 165–173.

Valentine, A J; B A Osborne and D T Mitchell (2001). Interactions between phosphorus supply and total nutrient availability on mycorrhizal colonization, growth and photosynthesis of cucumber. Sci. Hort., 88: 177–189.

Zhang, B; S Li; S Chen; T Ren; Z Yang; H Zhao and X Han (2016). Arbuscular mycorrhizal fungi regulate soil respiration and its response to precipitation change in a semiarid steppe. *Scientific reports*, *6*(1), 1-10.

Ziane, H; N Hamza and A Meddad-Hamza (2021). Arbuscular mycorrhizal fungi and fertilization rates optimize tomato (Solanum lycopersicum L.) growth and yield in a Mediterranean agroecosystem. *Journal of the Saudi Society of Agricultural Sciences*.

الملخص العربي

إستجابة نمو نباتات الجوافة والتنفسى الميكروبي بالتربة للتلقيح بأنواع الميكوريزا تحت ظروف الأراضي القلوية

هدى عبد الفتاح محمود[1]، ياسر ثابت عبد المجيد مصطفى [2]، وليد فضيلة عبد الجليل[3]

راغب محمد العادلى [1]،إسلام إبراهيم أحمد أبوالسعود[1]

1. قسم الارضى والكيمياء الزراعية – كلية الزراعة ساباباشا – جامعة الأسكندرية
2. قسم بحوث الليمنولوجى، المعمل المركزى لبحوث الثروة السمكية، مركز البحوث الزراعية.
3. قسم الإنتاج النباتى – كلية الزراعة ساباباشا – جامعة الأسكندرية

يهدف هذا البحث إلى اختبار تأثير أنواع فطر الميكوريزا *Rhizoglomus macrocarpium* (GM)*;* *Rhizoglomus irregularis* (GI) and *Rhizoglomus fasciculatum* (GF) ، في ظروف التربة القلوية ، على التنفس الميكروبي بالتربه من خلال انبعاث ثاني أكسيد الكربون ، في محاولة لزيادة صحة التربة وبالتالي تحسن نمو شتلات الجوافه. تم استخدام سماد النيتروجين (N) والبوتاسيوم (K) بمعدل 30 جم NH_4NO_3 و 20 جم K_2SO_4 لكل شتلة. تم وضع ثلاثة مستويات من السماد الفسفوري (سوبر فوسفات الكالسيوم (15.5٪ P_2O_5)) قبل الزراعه. تم تلقيح التربة بـ 30 مل من لقاح الميكوريزا قبل الزراعه بأسبوع. كما تمت إضافة 20 مل من لقاح الميكوريزا مع زراعه شتلات الجوافة. تم الري بماء الصنبور كل يوم. تم حصاد شتلات الجوافة بعد 120 يوم من الزراعة. أوضحت النتائج أن جميع أنواع الميكوريزا كانت فعالة في نمو شتلات الجوافه وامتصاص الفوسفور في شتلات الجوافة مقارنة بشتلات الجوافه الغير ملقحه. كانت اكثر انواع الميكوريزا تاثير علي نمو شتلات الجوافه وامتصاص الفوسفور (GI) *Rhizoglomus irregularis* حيث ادي إلى زيادة النشاط الميكروبي للتربة وهذا مدعوم بدليل على زيادة ثاني اكسيد الكربون، مما أدى إلى تحسين صحة التربة وزيادة توافر العناصر في التربة ، مما أدى إلى زيادة امتصاص الفوسفور بواسطة شتلات الجوافه.