**Research** Article

# Effects of arbuscular mycorrhizal fungi on enhancing growth, fruit quality, and functional substances in tomato fruits (*Lycopersicon esculentum* Mill.)

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Abstract This study aimed to investigate the efficiency of arbuscular mycorrhizal fungi (AMF) in enhancing plant performance and bioactive compound concentrations in tomatoes (Lycopersicon esculentum Mill.). This factorial pot experiment included nine replications over 120 days of cultivation. Three AMF species (Rhizophagus prolifer, *Claroideoglomus etunicatum*, and *Acaulospora mellea*) were utilized as inoculum, while non-mycorrhizal controls with or without synthetic NPK fertilizer were compared. Interestingly, C. etunicatum KS-02 inoculations effectuated the best fruit growth and weight, which were statistically higher than those of the control without AMF. However, only fruit fresh weight was higher in plants inoculated with C. etunicatum KS-02 than those treated with the synthetic NPK fertilizer. In addition, C. etunicatum KS-02 inoculations induced a  $\geq$  11% increase in DDPH (1,1-diphenyl-2-picrylhydrazyl) activity, lycopene content, and carotenoid content compared to the control. This study is the first to report Claroideoglomus species' effectiveness in promoting growth, fruit yield, and bioactive compound production in L. esculentum Mill. These findings substantiate the significant potential of C. etunicatum KS-02 for tomato cultivation without the adverse effects of excessive synthetic fertilizer use.

**Keywords** Antioxidants, Carotenoids, Lycopene, Phytochemical, Tomato production

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# Introduction

Tomato (Lycopersicon esculentum Mill.) is the second most important fruits and vegetable crop in the world, next to the potatoes. Ripe fruits are small, sweet and crispy in taste. They are commonly consumed as fresh fruit or can be eaten with salads. The fruits are rich in antioxidants such as carotenoids and vitamin C, with lycopene representing the main carotenoid, accounting for 80% of all carotenoids (Nguyen and Schwartz 1999). The fruits can be processed in a variety of industries to create products such as ketchup and tomato juice or used as an ingredient in some food condiments. This has resulted in a high demand value of up to 16 million tons per year. There is 6,328.8 hectare of tomato cultivation area in Thailand, most of this area being located in northeastern Thailand. The total yield in 2020 was 132,650 tons. However, tomato production in this area experiences many problems, including disease, insects or unacceptable weather conditions such as high temperatures, drought and low nutrient content in the soil. These factors result in lower yield and a poorer fruit quality of tomatoes (Lahoz et al. 2016). In light of this, a great deal of focus has fallen on new approaches to improve plant growth and increase specific substance content. At present synthetic fertilizer is commonly utilized to boost plant production (Seemakram et al. 2022). However, over long-term use, these synthetic fertilizers have been found to decrease soil quality with a notable reduction in beneficial soil microbes and evidence of toxic residues (Chandini et al. 2019). One way to lessen agriculture's dependency on synthetic fertilizers is through the use of plant-growthpromoting microorganisms (PGPM), which are a focus of interest and study because of their environmentally friendly nature (Andre et al. 2016; Seemakram et al. 2022).

Soil fertility has been declining as a result of cultivation

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for a long time. Thus, there is an evident need for cultivation management by other means, particularly with the soil biota, which facilitates improved growth performance of tomato fruit. Arbuscular mycorrhizal fungi (AMF), a soil biota belonging to the Division Glomeromycota, offers great potential. It involves a process of fungal symbiosis that colonizes the roots of over 80% of land plants (Boonlue et al. 2012) AMF symbiosis can be observed in nearly all ecosystems and occurs naturally in most plant species. One of the effects of this symbiotic relationship is to increase nutrient uptake from the soil for nutrients such as phosphorus (P), nitrogen (N) and potassium (K), and micronutrients that help for enhancing plant growth (Jeffries 1987). AMF also aids plant growth under adverse environmental conditions, such as aridity, and help prevent diseases in the root system (Seemakram et al. 2021). Moreover, AMF serves to help plants adjust the osmotic balance within their cells (Nacoon et al. 2021), and strengthen plant tolerance to stresses, such as salinity and heavy-metal pollution (Salam et al. 2017). Owing to the above benefits, some species of arbuscular mycorrhizal are of interest for utilization in bio-fertilizers to replace chemical fertilizers, etc.

Therefore, our principle objective was to investigate the role various species of AMF played on the growth performance, and the lycopene, carotenoid, and 1,1-diphenyl-2-picryl-hydrazyl (DPPH) content of the fruit of Lycopersicon esculentum Mill. varieties T72011. AMF treatment of the species was compared with two controls (without AMF inoculation and with chemical fertilizer treatment); a treatment with existing fertile soil and one with the addition of mineral fertilizer. However, there are currently no reports investigating the effects of AMF on the growth and combined production of secondary metabolites in tomatoes. The results obtained in this research could provide a framework for an in-situ application of AMF to increase tomato cultivation in Thailand. Mycorrhizal management offers economic benefits as an alternative to purchasing mineral fertilizer; therefore, this study offers potential options for the agricultural sector to decrease expenses and boost income.

# **Materials and Methods**

# AMF preparation

The AMF species, *Rhizophagus prolifer* PC2-2 (Seemakram et al. 2021, 2022), *Claroideoglomus etunicatum* KS-02 (Nacoon et al. 2023) and *Acaulospora mellea* KKU-NBP-SB-2 (Khaekhum et al. 2017), were obtained from Mycorrhiza and Mycotechnology Laboratory and used as the fungal

inoculum. The maize was used as the host plant for production of AMF spore following the methods of Boonlue et al. (2012). Soil was sterilized twice then placed in 8-inchdiameter plastic pots for use as a plant substrate. Surface sterilization of maize seed was performed by soaking in 6% sodium hypochlorite solution for 30 min. Around 200 AMF spores were added to the pots containing sterile maize seeds, before the maize was cultivated in a greenhouse at a temperature of 30-35°C with daily irrigation until 90 days, when watering ceased and the plants began to dry. The plants were cut just above the soil surface. Finally, soil samples were first dried and then crushed into finely ground particles. The purity of the spores and the total spore number were determined using the sucrose centri-

fugation method (Daniels and Skipper 1982). These dried

soils containing AMF spores, mycelia and infected roots were then used as the inoculum in the experiments.

# Soil preparation

The sandy loam soil properties include pH, electrical conductivity (EC), organic matter (OM), nitrogen (N), phosphorus (P), potassium (K), available phosphorus, exchangeable potassium, calcium (Ca), and sodium (Na) were analyzed according to the method described by Seemakram et al (2021). The soil samples were sterilized and stored overnight at room temperature, before being sterilized again under the same conditions, then packed in 6-inch diameter plastic pots; each pot contained 3 kg.

#### Tomato seed preparation

Tomato cultivars of *Lycopersicon esculentum* Mill. varieties T72011 were obtained by Asist. Prof. Dr. Chanon Lapjit, Faculty of Agriculture, Khon Kaen University, Thailand from the Horticultural station, Khon Kaen University, which develops and distributes them. The tomato seeds were sterilized with 6% sodium hypochlorite for 5 min, before being rinsed with sterile distilled water for 3 min. The sterilized seeds were placed in sterilized Petri dishes for 5 to 7 days, where root germination was observed. The seedlings were cultivated in peatmoss-filled trays for 2 weeks, after which the mature seedlings (of around 10cm in height) were transferred to plastic pots.

#### Experimental design and treatment

The pot experiment was carried out in a greenhouse at the Faculty of Science, Khon Kaen University, Thailand. The experimental design was conducted with a factorial completely randomized design (CRD) with 5 treatments in 6 replications as follows: (T1) control, sterilized soil without AMF; (T2) control with mineral fertilizer, sterilized soil without AMF with the addition of mineral nutrients (N,P,K: 15-15-15) at 0.32 g per pot; (T3) inoculation with *R. prolifer* PC2-2; (T4) inoculation with *A. mellea* KKU-NBP-SB-2; and (T5) inoculation with *C. etunicatum* KS-02. The AMF inoculum was applied adjacent to the plant root at a rate of approximately 200 spores/pot. After 3 months the plants were harvested.

#### Mycorrhizal root colonization

Mycorrhizal root colonization intensity was determined according to the method described by Koske and Gemma (1989). Roots were washed with 10% KOH for 3 min at 95°C, and then soaked in 2% HCl overnight. Trypan blue solution (0.05%) was used to stain root samples, which were then cut into 1 cm long pieces. The cellular structures of AMF, including vesicles, arbuscules, and hyphae, were observed using a microscope at 40 × magnification (SMZ745T Nikon, Japan) (Seemakram et al. 2022).

#### Determination of plant growth parameters

After 90 days of transplantation, the parameters of plant growth including plant height, stem diameter, SPAD and leaf area (Arnon 1949) were evaluated. The number and fresh weight of fruit were analyzed at harvest. Leaves, stem and roots were dried in an oven at 80°C for 3 days prior to analysis, and the dry weight measured to record plant biomass. The nutrient uptake concentration (N, P and K) of the samples was next determined. The parameters determining the quality of the plant roots, including the diameter, specific root length (SRL) and root tissue density (RTD), were measured by scanning the root samples using an Epson scanner V800 PHOTO, and the data were analyzed using WINRHIZO Pro2004a software (REGENT Instruments Inc., Quebec, QC, Canada).

#### Lycopene and Carotenoid content measurement

Fruit samples were cut and blended using a blender. Then, the samples were kept on ice in dark conditions. The sample of 0.2 g was transferred to a 50 mL covered test tube. A total of 20 mL of hexane: acetone: ethanol (HAE) of 2:1:1 (v/v/v) was added to the sample tubes. After that, the samples were mixed for 10 min and 3 mL of distilled water was added to each sample tube. The samples were agitated for several minutes. Then, the samples were kept

at room temperature. The supernatant of the hexane layer that contained lycopene and carotenoid was measured using a spectrophotometer at the wavelengths of 503 and 449 nm, respectively. The lycopene and carotenoid contents were calculated according to the formula described by Biswas et al. (2011) and Fester et al. (2002)

#### Antioxidant content measurement

Free-radical-scavenging activity of 1,1-diphenyl-2-picrylhydrazyl (DPPH) was detected modifying to the method described by Leong and Shui (2002). A freshly 0.1 mM solution of DPPH in methanol was prepared. A 100  $\mu$ L of each sample (with appropriate dilution) was first mixed with 4.0 mL of DPPH solution, before holding at room temperature for 30 min prior to measurement. The mixture solution was measured at 517 nm and detected by spectrophotometer. Methanol (0.5 mL), replacing the sample, was used as the blank. The percentage of radical-scavenging ability was analyzed by using the following formula:

Scavenging ability (%) = (Absorbance 517 nm of control - Absorbance 517 nm of sample) / Absorbance 517 nm of control × 100

#### Statistical analysis

All data from this work were evaluated by analysis of variance (ANOVA) for data from the factorial CRD. Fisher's least significant difference (LSD) was significant at  $p \leq 0.05$ . The correlation between data was analyzed using Statistix 10 software.

# Results

# Mycorrhizal colonization

The quantity of AMF spores and root colonization in *Lycopersicon esculentum* Mill. was measured and is reported in Table 1. In addition, AMF was found in the plant roots, such as hyphae, vesicles and arbuscules, in various structures. In the treatment without AMF (treatments T1 and T2), spores in the soils and tomato root colonization were not detected.

#### Effects of AMF on plant growth parameters

The ability of AMF to enhance plant growth was detected with respect to plant height, leaf greenness, diameter, leaf area, stem dry weight, leaf dry weight and root dry weight. In addition, the plant yield, including the number of fruits and the fruit fresh weight, was also evaluated (Table 2). The presence of AMF was found to increase all of the plant growth parameters in comparison to the control plant (T1). All of the plant growth parameters and yields in these treatments were significantly increased when compared to the control plant treatment. In the case of tomato inoculated with *C. etunicatum* KS-02 treatment, all plant growth parameters were found to be statistically higher than those of the control without AMF. Moreover, the highest yield or fruit fresh weight was also found in the *C. etunicatum* KS-02 treatment (T5). Therefore, the best AMF treatment for enhancing the growth of tomato was demonstrated to be *C. etunicatum* KS-02 (T5).

Table 3 shows the plant root growth performance of the tomato plants grown under different conditions. The results indicate that most of the root traits were significantly improved by inoculation with either AMF compared to control plant (T1). Nevertheless, the root dry weight and specific root length of the plants inoculated with *A. mellea* KKU-NBP-SB-2 (T4) and with *C. etunicatum* KS-02 (T5)

were significantly higher than those of the control plant (T1).

Effects of AMF on N, P and K contents

The effects of AMF on NPK concentration in tomato plants are indicated in Table 4. The tomato plants to which chemical fertilizer had been applied showed the maximum value of N.P. Among the AMF-inoculated treatments, N accumulation in the plants was increased but no significant difference was found with the non-inoculated control. The concentration of P in tomato plant inoculated with AMF was not statistically higher than that of the control treatment. However, the highest phosphorus content was detected in the tomato inoculation with R prolifer PC2-2. The concentration of potassium (K) was found to be statistically higher in all treatments inoculated with AMF than in the control. In the tomato plants inoculated with C. etunicatum KS-02, the highest K concentration was found. These results reveal that AMF enhanced mineral (N, P and K) contents in tomato plants, and particularly K, compared with the noninoculated control.

Table	1	AMF	spores	in	soil	and	AMF	colonization	in	tomato	roots
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Treatment	Spore/gram soil	Root colonization (%)
Tl	0.00c	0.00c
Τ2	0.00c	0.00c
Τ3	2.68b	20.19ab
Τ4	10.29ab	28.23a
<i>T5</i>	21.14a	29.01a
% cv.	11.36	28.09
F-test	*	n.s.

Numbers followed by the same letters in each column indicate values that were not significantly different according to the LSD test. \* significant difference at  $p \le 0.05$ , \*\* significant difference at  $p \le 0.01$ . T1: control without AMF inoculum; T2: chemical fertilizer; T3: *Rhizophagus prolifer* PC2-2; T4: *Acaulospora mellea* KKU-NBP-SB-2; T5: *Claroideoglomus etunicatum* KS-02. AMF, Arbuscular mycorrhizal fungi; n.s., not significant.

Treatment	SPAD	Height (cm)	Diameter (cm)	Leaf area (cm <sup>2</sup> )	Total number of fruits	Fruit fresh weight (g)	Stem dry weight (g)	Leaf dry weight (g)
<i>T1</i>	30.25b	68.80b	13.01b	15.09c	5b	10.16c	3.92b	1.96a
<i>T2</i>	45.32a	76.50ab	16.90a	30.78a	10a	18.38b	7.62a	2.18a
<i>T3</i>	40.18a	68.50b	15.10ab	25.75ab	8ab	24.16ab	5.56b	2.32a
T4	38.06a	74.50ab	13.70b	20.02b	11a	25.17ab	6.61ab	2.18a
<i>T5</i>	39.27a	78.60a	16.70a	29.35a	10a	40.70a	7.64a	2.67a
% cv.	20.18	25.01	24.50	27.52	24.31	29.07	27.09	25.67
F-test	*	**	**	*	**	*	*	n.s.

Table 2 Effects of AMF on tomato growth

T1: control without AMF inoculum; T2: chemical fertilizer; T3: *Rhizophagus prolifer* PC2-2; T4: *Acaulospora mellea* KKU-NBP-SB-2; T5: *Claroideoglomus etunicatum* KS-02. AMF, Arbuscular mycorrhizal fungi; n.s., not significant.

Effects of AMF on antioxidant activity, lycopene content and carotenoid content

Fruit lycopene content, carotenoid content and antioxidant activity increased with all of the AMF treatments compared to the control treatment without fertilizer, and no significant difference was found with chemical fertilizer treatment. The maximum antioxidant activity (97.29%), lycopene content (0.62  $\mu$ g/g) and carotenoid content (3.22  $\mu$ g/g) were found in the *C. etunicatum* KS-02 treatment, which showed a

significant difference from the non-inoculated control (Table 5). In addition, this finding was not significantly different to chemical fertilizer treatment. Therefore, the best performance in terms of fruit lycopene content, carotenoid content and antioxidant activity was found in the treatment of the plant inoculated with *C. etunicatum* KS-02, which might be used instead of chemical fertilizer.

Correlations between AMF, % colonization, nutrient uptake and the concentration of secondary compounds and the plant growth parameters of tomato at the harvest stage

Table 3 Effects of AMF on tomato root growth

Treatment	Root dry weight (g)	Average diameter (mm)	Specific root length $(m g^{-1})$	Root tissue density (g cm <sup>-3</sup> )
<i>T1</i>	0.91ab	0.28a	107.95b	94.02a
<i>T2</i>	1.23a	0.30a	113.31ab	78.03ab
<i>T3</i>	0.88b	0.29a	113.18ab	83.60ab
<i>T4</i>	1.09ab	0.28a	124.11ab	81.78ab
<i>T5</i>	1.24a	0.28a	149.93a	67.69b
% cv.	21.36	28.09	25.71	24.02
F-test	*	n.s.	*	*

T1: control without AMF inoculum; T2: chemical fertilizer; T3: *Rhizophagus prolifer* PC2-2; T4: *Acaulospora mellea* KKU-NBP-SB-2; T5: *Claroideoglomus etunicatum* KS-02. AMF, Arbuscular mycorrhizal fungi; n.s., not significant.

Treatment	N concentration (mg g <sup>-1</sup> )	P concentration (mg g <sup>-1</sup> )	K concentration (mg g <sup>-1</sup> )
<i>T1</i>	16.6b	10.2b	10.9b
T2	21.2a	23.3a	13.7ab
<i>T3</i>	17.5ab	15.1ab	15.7a
Τ4	16.9b	10.1b	14.4ab
<i>T5</i>	19.9ab	13.8b	15.9a
% cv.	29.09	25.83	27.42
<i>F-test</i>	*	*	*

Table 4 Effects of AMF on nitrogen, phosphorus, and potassium concentrations in tomatoes

T1: control without AMF inoculum; T2: chemical fertilizer; T3: *Rhizophagus prolifer* PC2-2; T4: *Acaulospora mellea* KKU-NBP-SB-2; T5: *Claroideoglomus etunicatum* KS-02. AMF, Arbuscular mycorrhizal fungi; N, Nitrogen; P, Phosphorus; K, Potassium.

Table 5 Effect of AMF on antioxidant activity, lycopene content, and carotenoid content in tomatoes

Treatment	DPPH (%)	Lycopene (µg/g)	Carotenoid (µg/g)
Τ1	87.14b	0.42b	2.37b
<i>T2</i>	96.26a	0.52ab	3.38a
<i>T3</i>	96.23a	0.49b	3.26a
T4	95.61a	0.59ab	3.16a
<i>T5</i>	97.29a	0.62a	3.22a
% cv.	27.09	25.71	25.32
F-test	*	*	*

T1: control without AMF inoculum; T2: chemical fertilizer; T3: *Rhizophagus prolifer* PC2-2; T4: *Acaulospora mellea* KKU-NBP-SB-2; T5: *Claroideoglomus etunicatum* KS-02. AMF, Arbuscular mycorrhizal fungi.

are shown in Table 6. Mycorrhizal colonization and root dry weight significantly positively correlated with the number of fruit, fruit fresh weight, antioxidant activity, lycopene content and K concentration of the plant. Among the AMF treatments, *C. etunicatum* KS-02 was significantly positively correlated with fruit fresh weight, antioxidant activity, lycopene content and the K concentration of the plant. The number of fruit and fruit fresh weight were significantly

number of fruit and fruit fresh weight were significantly positively correlated with DPPH activity, lycopene content and carotenoid content, and the K concentration of the plant. Additionally, DPPH activity, lycopene content and carotenoid content were significantly positively correlated with the K concentration of the plant.

# Discussion

According to our previous studies, the species of AMF showed high efficiency in enhancing plant growth and biomass in perennial plants belonging to the legume family (Siamese rosewood and Burma padauk) (Seemakram et al. 2021) and cannabis (Seemakram et al. 2022). Therefore, we expected inoculation with AMF to play a key role in boosting the growth and production of secondary metabolites in tomato. The efficiency of three AMF species, namely *R. prolifer* PC2-2, *C. etunicatum* KS-02 and *A. mellea* KKU-NBP-SB-2, was investigated for its capacity to promote the growth and production of secondary meta-

bolites of tomato (*L. esculentum* Mill.), compared with the effect of the application of synthetic NPK fertilizer under greenhouse conditions. *Claroideoglomus etunicatum* KS-02 showed a higher root colonization than that of *A. mellea* KKU-NBP-SB-2 (28.23%) and *R. prolifer* PC2-2 (20.19%). A study using concentrations of *C. etunicatum* up to 51% (Ziane et al. 2017) observed higher root colonization in tomato roots compared with our findings. The data from our study is similar to the value of the mycorrhizal colonization of black rice (Maled Phai and Niew Dam Hmong) of 25, and the value of 9% reported by Nacoon et al. (2023).

This study identified that colonization with AMF affected growth performance of tomato cultivars and the production of secondary metabolites of tomato fruits beneficially under greenhouse conditions. Treatment with C. etunicatum KS-02 significantly affected the SPAD value, height, diameter, leaf area and biomass of the tomato plant. Our data show similar results to those of Ziane et al. (2017), who reported that the overall height and biomass of tomato inoculated with the commercial AMF inoculum were significantly increased. Mutumba et al. (2018) documented the effect of mycorrhizal fungi on improving the plant growth parameters and chlorophyll index of the host plant. In addition, our results concur with those in previous literature using other plant species. Inoculation with R. prolifer PC2-2 in Siamese rosewood, Burma padauk and cannabis resulted in an increase in the leaf area and biomass (Seemakram et al. 2021).

Table 6 Correlation between AMF, nutrient uptake, antioxidants, lycopene, and carotenoids in tomatoes

Correlation	CF	AMF1	AMF2	AMF3	Root coloni- zation	No. of spores	Root dry weight	No. of fruit	Fruit fresh weight	DPPH	Lyco- pene	Caro- tenoids	Ν	Р
Root colonization	-0.59**	0.18 ns	0.48*	0.51*										
No. of spores	-0.42 ns	-0.25 ns	s 0.21 ns	0.88**	0.81**									
Root dry weight	0.52*	-0.62**	0.06 ns	0.55*	0.16 ns	0.53*								
No. of fruit	0.28 ns	-0.18 ns	s 0.51*	0.28 ns	0.58*	0.53*	0.73**							
Fruit fresh weight	-0.26 ns	0.02 ns	0.07 ns	0.84**	0.83**	0.92**	0.54*	0.63**						
DPPH	0.23 ns	0.23 ns	0.14 ns	0.37 ns	0.60**	0.49*	0.57*	0.86**	0.75**					
Lycopene	0.10 ns	-0.01 ns	s 0.36 ns	0.47*	0.73**	0.68**	0.67**	0.94**	0.82**	0.94**				
Carotenoids	0.41 ns	0.25 ns	0.11 ns	0.19 ns	0.43 ns	0.29 ns	0.55*	0.84**	0.58*	0.97**	0.87**			
N	0.76**	-0.25 ns	<b>-0.41ns</b>	0.40 ns	-0.15 ns	0.18 ns	0.81**	0.48 ns	0.35 ns	0.56*	0.47*	0.62**		
Р	0.91**	0.06 ns	-0.45**	-0.07 ns	-0.42 ns	-0.29 ns	0.48 ns	0.30 ns	-0.02 ns	0.46 ns	0.24 ns	0.61**	0.85**	
K	-0.11 ns	0.43 ns	0.07 ns	0 49*	0 77**	0.61**	033 ns	0 68**	0 85**	0 92**	0 86**	0 83**	033 ns (	) 19 ns

\*\*, significant difference at  $p \le 0.01$ ; \*, significant difference at  $p \le 0.05$ ; ns, not significant. *R. prolifer* PC2-2 (AMF1), *A. mellea* KKU-NBP-SB-2 (AMF2); and *C. etunicatum* KS-02 (AMF3)

Moreover, black rice inoculated with C. etunicatum outperformed non-inoculated plants in the absence of mineral fertilizer in terms of biomass (Nacoon et al. 2023). Mineral fertilizer boosted plant performance when applied, but the effect was non-significant in terms of biomass. The symbiotic relationship between AMF and host plants enhances plant growth and biomass, and enables the plant to resist abiotic stress conditions (Alam et al. 2023). Furthermore, AMF inoculation significantly affected the number of tomato fruits yielded and fruit fresh weight. Consistent with previous studies, the incorporation of inoculated Funneliformis *mosseae* into the continuous cropping substrate remarkably improved the growth and yield of tomatoes, and resulted in a slender rise in fruit size in tomato production (Wang et al. 2021). The above results affirm that AMF inoculation resulted in greater fruit numbers regardless of the fertilization criterion in tomato.

As may be expected, for the roots of tomato plants colonized by AMF in all treatments, the root qualities were affected. However, the root dry weight and specific root length after treatment with C. etunicatum KS-02 were significantly higher than those of non-inoculated plants. This may have resulted from the introduction of AMF, leading to more effective root colonization, which affected the root qualities, especially root dry weights, which were significantly greater in inoculated plants compared to non-inoculated plants (Ziane et al. 2017). These results suggest that the inoculation of AMF had an effect on specific root traits, but not all of them (Seemakram et al., 2022). Furthermore, plants inoculated with C. etunicatum KS-02 exhibited slightly higher N, P and K concentrations. This is perhaps due to mycorrhizal plants exploring a greater volume of soil for available nutrients and water than uninoculated ones, leading to the recorded increase in N, P and K (Evelin et al. 2012). However, considering nutrient concentration among the tomato plants, all plants inoculated with AMF demonstrated a value of K which was significantly higher than that of the uninoculated plant.

Both inoculation with AMF and synthetic fertilizer served to increase the performance of tomato and increased the capacity of the DPPH, lycopene content and carotenoid content compared to the unfertilized control. The maximum DDPH activity and lycopene and carotenoid contents for the *C. etunicatum* KS-02 treatment were found to be higher than those seen in the non-inoculated control treatment. A positive correlation was found between K uptake and an increase in DPPH activity, in addition to lycopene and carotenoid content. Moreover, inoculation with *C. etunicatum* KS-02 in tomato cultivation showed a positive correlation between lycopene and K uptake. Lycopene is one of the precursors of carotenoid synthesis, formed through the cyclization of lycopene, which has a positive correlation with K (Heldt 2003; Taber 2006). In addition, root colonization and root dry weight exhibited positive correlations with the number of fruit and fruit fresh weight, resulting in increased lycopene and carotenoid accumulation in the fruit. Our data are similar to those of Ordookhani et al. (2010), who reported that a positive correlation between lycopene and shoot potassium was found in plants treated with PGPR. Stimulation of the carotenoid metabolism was correlated with the root colonization of AMF (Fester et al. 2002). AMF colonization activated plant defense mechanisms producing of phenolics and flavonoids (Zhao et al. 2022). Avio et al. (2017) reported that R. irregulare IMA6 increased the phenolics concentration and antioxidant activity in tomato plants. In general, tomato cultivation with AMF led to increased carotenoid and total phenolic contents, while AMF inoculation also increased lycopene content in cultivated tomatoes (Ulrichs et al. 2008). In previous work reported that the increase in functional substances in plants was correlated with increases in phosphorus acquisition (Seemakram et al. 2022). Our results contradict those from previous studies, which partly supported the nutritional mechanism of tomato, indicated by the correlations between AMF and K concentration with an increase in antioxidant activity, lycopene content and carotenoid content.

Our study shows that using AMF obtained a similar result in terms of plant growth performance, tomato yield and functional compounds in tomato fruit compared with the application of chemical fertilizer; inoculation with C. etunicatum KS-02 was particularly successful. Further, high levels of antioxidant compounds, such as lycopene and carotenoids, in tomato fruit may boost market price, especially where the tomato plant is cultivated organically (Pataro et al., 2020). Nevertheless, the use of AMF alone had substantial benefits regarding plant growth and quality and functional compounds in tomato fruit. As highlighted above, the use of chemical fertilizers enhanced some physical plant characteristics more rapidly than with the use of AMF. Best practice for agribusiness would be utilizing chemical fertilizers in combination with biological techniques in order to achieve the highest efficiency in the production of tomato yield and to maximize the quality of fruit with a high content of functional compounds, while minimizing environmental damage. In this regard, the inoculation of C. etunicatum KS-02 could, with careful management, help farmers to reduce the production costs of tomato cultivation by providing a partial or complete alternative to chemical fertilizer. The combined use of AMF and fertilizer application, including organic soil management, is the main target that warrants further investigation.

## Conclusions

Our study demonstrates the effect of AMF inoculation on the growth promotion, yield and bioactive compound production of tomato (*L. esculentum* Mill.), showing no significant difference from chemical fertilizer application. AMF treatment also led to significant correlations between mycorrhizal colonization, fruit weight, fruit number, concentration of K and bioactive compound content. We found that *Claroideoglomus etunicatum* KS-02 was the best plant growth promoter, being able to promote growth, fruit yield, antioxidant capacity, lycopene and carotenoids in tomato compared with noninoculated plants under unfertilized conditions. This species warrants further analysis as a means to developing an AMF inoculum alternative for the industrial production of tomato, replacing the immoderate application of synthetic fertilizer which characterizes current conventional practices.

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