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Interaction Between AMF and Plant Growth-Promoting Rhizobacteria on Two Varieties of *Solanum lycopersicum* L.

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Abstract: Arbuscular mycorrhizal fungi (AMF) and bacteria can interact synergistically to stimulate plant growth, despite abundant amount of phosphorus in parent material, the soil phosphorus availability is limited for plant. This study investigated the interaction of phosphate solubilizing bacteria (PSB) and Arbuscular mycorrhizal fungi (AMF) on two verities of *Solanum lycopersicum* L(Var, NS 524 and NS 585). The experimental design was split plot factorial with on a complete randomized block design. The treatments included soil: sand (v/v 3:1) two levels of phosphate solubilizing micro-organisms. The growth parameters like root length, fresh weight of root, dry weight of root, fresh weight of shoot, dry weight of shoot, percentage root colonization, spore number and number of leaves, were significantly increased by inoculating *with Glomus fasciculatum, Pseudomonas fluorescens* and *Azotobacter chrococcum* and enhanced tomato shoot and root biomass.

Key words: *Pseudomonas fluorescens* · *Solanum lycopersicum* L. · Root length · Spore number

INTRODUCTION

In the last century, chemical fertilizers were introduced and this made farmers to be happy of getting increased yield in agriculture in the beginning. But slowly chemical fertilizer started displaying their ill-effects such as leaching, polluting water basins, destroying microorganisms and friendly insects, making the crop more susceptible to the attack of diseases, reducing the soil fertility and thus causing irreparable damage to the overall system. plant growth promoting rhizobacteria (PGPR) are a group of bacteria that actively colonize plant roots and increase plant growth and yield [1,2,3]. Plant growth benefits due to the addition of PGPR include increase in germination rates, root growth, shoot and root weights [4], grain yield [5,6], chlorophyll content [7] tolerance to drought, salt stress and delayed leaf senescence [8]. PGPR increase the growth of a number of important crops, with some strains inducing systemic resistance to fungi, bacteria viruses and in some cases nematodes [9,10]. In addition to these effects of bacteria on AM fungi, the AM fungi themselves have also been shown to have an impact on the composition of bacterial communities [11]. The facts that, the dual inoculation of AM and specific PGPR can enhance the activity of AM during the symbiosis with the host plant [12,13,14]. In addition to these effects of bacteria on AM fungi, the AM fungi themselves have also been shown to have an impact on the composition of bacterial communities [11]. This impact may be relayed through the plant root because mycorrhizal establishment has been shown to change the chemical composition of root exudates and these are often a source of nutrients to associated bacteria in the mycorrhizosphere [15-21]. Finding the microorganisms very close to epidermis, plants secrete signal molecules for protection against invasion of the heterogeneous microbes in the root zone and at this stage the differentiation takes place between pathogenic, associative, symbiotic, or neutralistic adaptation of microbes with the plant [22]. In this article, we will focus on interactions between bacteria and AM fungi with proven or potentially synergistic properties that lead to stimulation of plant growth.

MATERIALS AND METHODS

Soil Plant Material and Bioinoculant Selection: The collection of AM fungi, steam sterilization procedure of

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soil, collection of seeds, seed sterilization and pots used for experiment, steam sterilized soil was filled to pots. The *Azotobacter chroococcum* (Ac) ACD 15 and *Pseudomonas fluorescens* (Pf) WGUK 327 were collected from microbiology laboratory, University of Agricultural sciences Dharwad, India. Seeds were sown in pots and after germination uniform seedlings were made one per pot.

Inoculation of AM Fungi: The *Glomus fasciculatum* (Thaxter) Gerdmann and Trappe emend. Walker & Koske., were mass multiplied in 35 cm diameter containing 8.5 kg using sterilized sand: soil (1:1 v/v) mixture as the substrate and (*Sorghum vulgare* L.) Jowar as the host. After 60 days of growth shoots of Jowar were chopped and the inoculum containing spores, root bits was air dried log of the mycorrhizal inoculum was applied to the planting area at a depth of about 4 cm to the pots (except non-inoculated control) before sowing seeds.

Experimental Design: The experiment was completely randomized with three replications of each treatment 3ml of culture suspension containing 10^{10} cells/ml of *Azotobacter chrococcum* (Ac) ACD 15 and *Pseudomonas fluorescens* (Pf) WGUK 327 in the form of slurry was inoculated on the inoculum as AMF+Ac and AMF+Pf and in triple inoculation AMF+Ac+Pf was inoculated. A non-inoculated control without bio-inoculants was maintained. The treatments were as follows NS 524 and NS 585 varieties of *Solanum lycopersicum* L.

- Noninoculated control
- Glomus fasciculatum
- Gf+Ac
- Gf+Pf
- Gf+Ac+Pf

The plants were exposed to sunlight and were kept free of weeds and irrigated properly. The plants were harvested after 30, 60 and 90 days. After 90 days seeds number and weight were recorded. The percentage of mycorrhizal infection was evaluated microscopically followed by clearing of roots in 10 % KOH neutralized in 2% HCL and stained with 0.05% trypan blue in lactophenol according to the method described by [23] and oot colonization was calculated as mentioned below.

$$\text{Percent of root colonization (\%)} = \frac{\text{No of root bits colonization}}{\text{Total number of root bits observed}} \times 100$$

The growth parameters like Shoot length, fresh weight of shoot, dry weight of root, dry weight of root, number of leaves, number of flowers and number of fruits, shoot and dry weight were determined after drying the plant samples in a hot air oven. The AM fungal spores were counted in 50 g of soil by wet sieving and decanting [24]. The phosphorus content in the shoots in terms of percentage was determined by the Vanadomolybdate phosphoric yellow colour method in plant sample [25].

Statistical Analysis: The data were statistically analyzed using analysis of variance (ANOVA) with the help of the SPSS software on computer. The mean values were compared by Duncan's multiple range tests at 0.05 level of significance.

RESULTS

The mycorrhizal inoculation (*Glomus fasciculatum*) with two plant growth promoting rhizobacteria (*Azotobacter chrococcum* and *Pseudomonas fluorescens*) increased significantly the height, shoot and root biomass of NS 524 and NS 585 varieties (Table 1 and 2) as compared to non mycorrhizal control plants. The positive fungal effect on *Solanum lycopersicum* L., was modified when *Azotobacter chrococcum* and *Pseudomonas fluorescens* was added together with the fungus.

The effect of *Azotobacter chrococcum* and *Pseudomonas fluorescens* with mycorrhiza on NS 524 varieties of *Solanum lycopersicum* L., significantly corresponds to better growth. After 30 days of plant growth there was a significant increase in fresh(9.45g) and dry weight of shoot(1.25g), root length(12.54cm), fresh(1.85g) and dry weight of root(0.28g), number of leaves(36.33), root colonization(69.3%), phosphorus content (0.24%), number of spores(71.00), in root inoculated with *Glomus fasciculatum, Azotobacter chrococcum* and *Pseudomonas fluorescens.* There were no significant records of number of fruits and flowers during this stage. The plant growth after 60 days in the triple inoculation with *Glomus fasciculatum, Azotobacter chrococcum* and *Pseudomonas fluorescens* increased significantly and recorded shoot length (78.29cm), fresh (133.59g) and dry weight of shoot (16.43g), root length (38.33g), fresh (6.19g) and dry weight of root(1.87g), even in number of flowers(12.33), fruits (5.33), there were significant increase inoculated with *Glomus fasciculatum, Azotobacter chrococcum* and *Pseudomonas fluorescens*

Table 1: Showing effect of *Glomus fasciculatum,* on growth characteristics of *Solanum lycopersicum* L. var. NS 524 for 30, 60 and 90 days.SL-shoot length, FWS-fresh weight of shoot,DWS-Dry weight of shoot,RL-Root length, FWR-Fresh weight of root,DWR-Dry weight of root, NL-Number of leaves,NF_wNumber of flowers, NF_r- Number of fruits, PC-percent of root colonization, SN-Spore number, SD-Stem diameter, P-uptake-Phosphorus

Treatments	SL(cm)	FWS(gm)	DWS(gm)	RL(cm)	FWR(gm)	DWR(gm)	NL	NF_w	NF_r	PC(%)	SN	SD(cm)	P uptake(%)
30 days													
CN	8.35±0.18e	3.37±0.13d	0.18±0.05e	6.36±0.25d	0.42±0.05d	0.03±0.05d	11.33±3.66c	0.00±0.00e	0.00±0.00e	0.00±0.00e	0.00±0.00e	1.00±0.05c	0.08±0.03d
Gf	16.28±0.15b	7.38±0.11b	0.94±0.01b	9.30±0.18b	0.86±0.01b	0.08±0.03b	25.00±0.57b	0.00±0.00d	0.00±0.00d	60.76±0.24b	74.62±0.09b	1.51±0.08b	0.13±0.03b
Gf+ Az	12.35±0.18d	5.44±0.10c	0.34±0.01d	7.52±0.01c	0.65±0.01c	0.05±0.33c	21.00±0.57b	0.00±0.00c	0.00±0.00c	45.68±0.12d	65.48±0.07d	0.96±0.03c	0.12±0.00c
Gf+Pf	13.43±0.22c	5.78±0.19c	0.38±0.05c	7.83±0.01c	0.65±0.01c	0.07±0.03b	22.66±0.33b	0.00±0.00b	0.00±0.00b	58.63±0.06c	70.46±0.08c	1.54±0.01b	0.13±0.00b
Gf+Az+Pf	20.37±0.19a	9.45±0.01a	1.25±0.05a	12.54±0.08a	1.85±0.05a	0.28±0.05a	36.33±0.33a	0.00±0.00a	0.00±0.00a	70.35±0.19a	85.95±0.12a	1.85±0.05a	0.24±0.05a
60 days													
CN	51.18±0.09e	48.93±0.01e	3.57±0.13e	18.00±0.57d	3.06±0.01e	1.44±0.02c	65.33±0.33e	3.33±0.33c	1.66±0.33c	0.00±0.00e	0.00±0.00e	1.96±0.03d	0.09±0.03d
Gf	66.26±0.15c	80.31±0.02b	12.48±0.02b	25.33±0.33b	5.37±0.01b	1.77±0.08b	85.33±0.33b	4.33±0.33c	4.66±0.33a	63.18±0.10b	64.66±0.33b	3.50±0.06b	0.16±0.03b
Gf+ Az	62.42±0.22d	53.38±0.26d	4.91±0.04d	22.33±0.33c	4.82±0.01c	1.44±0.01c	78.00±0.57d	6.66±0.33b	3.33±0.33b	56.24±0.12d	50.33±0.33d	2.96±0.06c	0.13±0.03c
Gf+Pf	72.36±0.18b	77.65±0.07c	11.07±0.05c	22.00±0.57c	4.38±0.05d	1.43±0.01c	81.00±0.57c	12.33±0.33a	4.66±0.33a	59.28±0.14c	57.66±0.33c	3.00±0.03c	0.14±0.03c
Gf+Az+Pf	78.29±0.14a	133.59±0.18a	16.43±0.04a	38.33±0.33a	6.19±0.05a	1.87±0.08a	91.66±0.88a	12.33±0.66a	5.33±0.33a	78.32±0.16a	88.66±0.33a	3.85±0.02a	0.29±0.01a
90 days													
CN	58.32±0.21e	49.24±0.08e	9.27±0.14e	20.33±0.33e	7.44±0.02e	1.86±0.01e	78.66±0.33e	2.66±0.33c	3.66±0.33d	0.00±0.00e	0.00±0.00e	2.61±0.08d	0.11±0.03e
Gf	76.46±0.24b	92.50±0.07b	16.81±0.03b	41.00±0.57b	12.59±0.05b	3.27±0.08b	95.00±0.57b	4.33±0.33b	6.66±0.33b	73.47±0.25b	75.23±0.13b	3.53±0.01b	0.20±0.05b
Gf+ Az	62.41±0.21d	65.77±0.01d	10.23±0.10d	35.33±0.33d	10.56±0.11d	2.83±0.01d	81.33±0.33d	4.66±0.33	5.33±0.33c	60.23±0.13d	63.42±0.22d	3.00±0.06d	0.15±0.03d
Gf+Pf	64.47±0.24c	82.53±0.23c	13.55±0.01c	38.66±0.33c	11.41±0.16c	2.96±0.08c	84.66±0.33c	3.66±0.33b	2.66±0.33d	65.53±0.26c	70.51±0.26c	3.13±0.07c	0.17±0.03c
Gf+Az+Pf	89.15±0.15a	112.48±0.20a	19.77±0.10a	44.66±0.33a	15.94±0.02a	7.75±0.01a	113.00±0.57a	5.66±0.33a	10.00±0.57a	90.39±0.21a	103.33±0.17	4.47±0.24a	0.38±0.03a

CN-Control, Gf-*Glomus fasciculatum*; *Glomus fasciculatum+ Azotobacter chrococcum + Pseudomonas fluorescens*. Mean values followed by the same letter are not significantly different at P=0.05. Mean ± Standard error.

Table 2: Showing effect of *Glomus fasciculatum,* on growth characteristics, root colonization, spore number of *Solanum lycopersicum* L. var. NS 585 for 30, 60 and 90 days. SL-shoot length, FWS-fresh weight of shoot,DWS-Dry weight of shoot,RL-Root length, FWR-Fresh weight of root,DWR-Dry weight of root, NL-Number of leaves,NF_wNumber of flowers, NF_r- Number of fruits, PC-percent of root colonization, SN-Spore number, SD-Stem diameter, P-uptake-Phosphorus

Treatments	SL(cm)	FWS(gm)	DWS(gm)	RL(cm)	FWR(gm)	DWR(gm)	NL	NF_w	NF_r	PC(%)	SN	SD(cm)	P uptake (%)
30 days													
CN	9.08±0.04e	9.15±0.01e	0.65±0.03e	1.22±0.01d	0.95±0.01e	0.12±0.02d	23.33±0.33e	0.00±0.00e	0.00±0.00e	0.00±0.00c	0.00±0.00e	1.11±0.05	0.03±0..05d
Gf	30.40±0.20c	8.37±0.05c	0.84±0.05b	13.29±0.18b	0.93±0.08b	0.12±0.05b	42.66±0.33b	0.00±0.00d	0.00±0.00d	58.81±3.41b	68.00±0.57b	2.53±0.01b	0.08±0.03b
Gf+ Az	28.30±0.15d	2.86±0.01d	0.12±0.08d	10.24±0.12d	0.64±0.05d	0.08±0.03c	22.00±0.33d	0.00±0.00c	0.00±0.00c	58.35±0.18b	58.33±0.33d	3.00±0.05a	0.14±0.03c
Gf+Pf	32.21±0.14b	10.54±0.02b	0.71±0.05c	12.27±0.13c	0.86±0.01c	0.11±0.05b	35.00±0.57c	0.00±0.00b	0.00±0.00b	60.43±0.24b	60.33±0.33c	2.55±0.02b	0.11±0.03b
Gf+Az+Pf	35.20±0.14a	13.06±0.02a	1.22±0.01a	15.24±0.13a	1.07±0.05a	0.28±0.03a	45.33±0.33a	0.00±0.00a	0.00±0.00a	78.36±0.18a	78.33±0.33a	3.10±0.05a	0.15±0.03a
60 days													
CN	69.09±0.06e	69.09±0.06e	5.71±0.02e	22.18±0.11e	4.16±0.05e	0.77±0.08e	72.33±0.33e	5.66±0.33c	1.66±0.33d	0.00±0.00e	0.00±0.00e	2.02±0.03c	0.01±0.05e
Gf	83.46±0.05b	83.46±0.05b	9.59±0.03b	42.23±0.16b	5.86±0.01b	1.54±0.08b	92.33±0.33b	7.60±0.33b	3.33±0.33c	69.34±0.17b	74.66±0.33c	3.09±0.04b	0.18±0.03c
Gf+ Az	70.74±0.09d	70.74±0.09d	6.38±0.05d	25.24±0.13d	4.95±0.01d	1.16±0.01d	81.00±0.57d	6.66±0.33bc	3.00±0.00c	55.37±0.19d	70.66±0.33d	3.17±0.04b	0.15±0.03d
Gf+Pf	72.76±0.08c	72.76±0.08c	7.25±0.02c	35.40±0.22c	5.56±0.01c	1.42±0.05c	84.66±0.33c	7.33±0.33b	4.33±0.33b	65.42±0.22c	78.33±0.33b	3.23±0.03b	0.20±0.03b
Gf+Az+Pf	92.54±0.02a	92.54±0.02a	11.55±0.01a	49.47±0.25a	10.25±0.01a	2.56±0.01a	103.66±0.20a	10.00±0.57a	5.66±0.33a	91.40±0.21a	99.00±0.57a	3.68±0.14a	0.22±0.05a
90 days													
CN	49.24±0.08e	49.24±0.08e	9.27±0.14e	20.33±0.33e	7.44±0.02e	1.86±0.01e	78.66±0.33e	2.66±0.33c	3.66±0.33d	0.00±0.00e	0.00±0.00e	2.61±0.08d	0.12±0.03d
Gf	92.50±0.07b	92.50±0.07b	16.81±0.03b	41.00±0.57b	12.59±0.05b	3.27±0.08b	95.00±0.57b	4.33±0.33b	6.66±0.33b	73.47±0.25b	75.23±0.13b	3.53±0.01b	0.28±0.03b
Gf+ Az	65.77±0.01d	65.77±0.01d	10.23±0.10d	35.33±0.33d	10.56±0.11d	2.83±0.01d	81.33±0.33d	5.66±0.33a	5.33±0.33c	60.23±0.13d	63.42±0.22d	3.00±0.06cd	0.23±0.06c
Gf+Pf	82.53±0.23c	82.53±0.23c	13.55±001c	38.66±0.33c	11.41±0.16c	2.96±0.08c	84.66±0.33c	3.66±0.33bc	2.66±0.33d	65.53±0.26c	70.51±0.26c	3.13±0.07c	0.24±0.03c
Gf+Az+Pf	112.48±0.20a	112.48±0.20a	19.77±0.10a	44.66±0.33a	15.94±0.02a	7.75±0.01a	113.00±0.57a	4.33±0.33b	10.00±0.57a	90.39±0.21a	103.33±0.17a	4.47±0.24a	0.42±0.08a

CN-Control, Gf-*Glomus fasciculatum*; *Glomus fasciculatum+ Azotobacter chrococcum + Pseudomonas fluorescens*.Mean values followed by the same letter are not significantly different at P=0.05. Mean ± Standard error

compared to other treatments. The root colonization recorded (78.32%), spore number (88.66) in 50g of soil and phosphorus content (0.29%) in root inoculated with *Glomus fasciculatum, Azotobacter chrococcum* and *Pseudomonas fluorescens* than dual inoculation and single inoculation. After 90 day, the plants growth responded in the similar trend inoculated with *Glomus fasciculatum, Azotobacter chrococcum* and *Pseudomonas fluorescens* and the recorded values are, shoot length (89.15cm), fresh (112.48g) and dry weight of shoot (19.77g), root length (44.66 cm), fresh (15.94) and dry weight of root (7.75g) and number of leaves (113.00) were recorded significantly in the inoculated, *Glomus fasciculatum, Azotobacter chrococcum* and *Pseudomonas fluorescens* (Table 1). The triple inoculation resulted largest increase in number of fruits (10.00), root colonization(90.39%), spore number(103.33) in 50 g of soil, phosphorus content in root (0.38%). In dual inoculation of *Glomus fasciculatum* and *Pseudomonas fluorescens,* resulted highest when compared with *Glomus fasciculatum* and *Azotobacter chrococcum*. There was no significant record of flowers during this stage. The dry weight of root and phosphorous uptake (Fig. 2 a, c).

The inoculation of *Glomus fasciculatum, Azotobacter chrococcum* and *Pseudomonas fluorescens* on NS 585 varieties of *Solanum lycopersicum* L., significantly increased the extent of AM colonization of the root system (Fig. 3 b). After 30 days responded

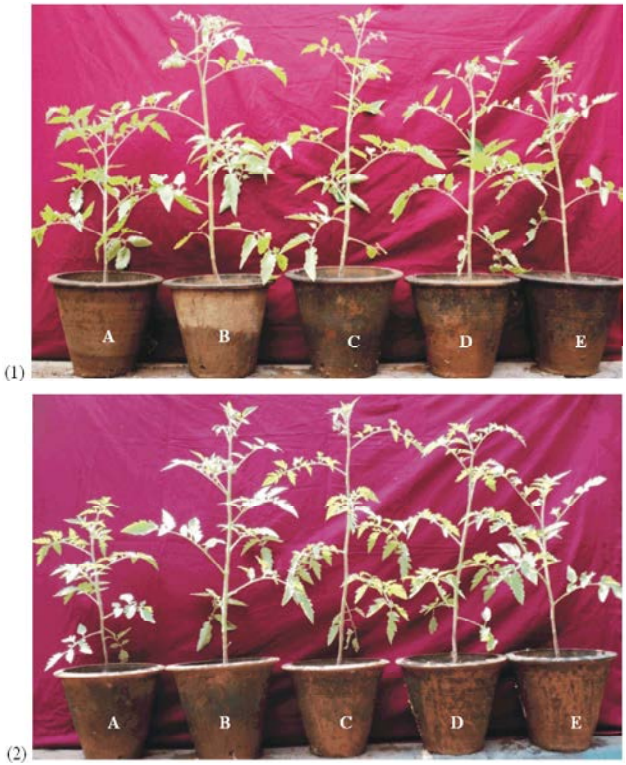


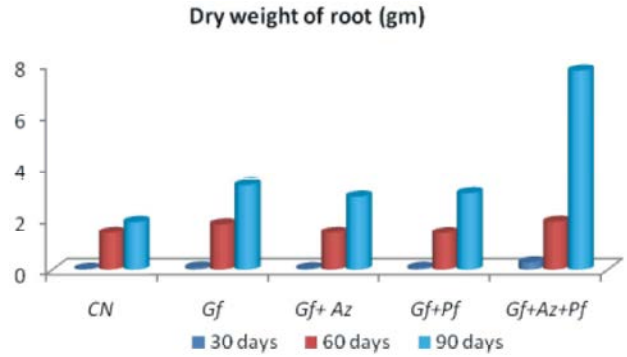
Fig. 1: Showing symbiotic response of Non inoculated control, *Glomus fasciculatum,* Gf+Ac, Gf+Pf, Gf+Ac+Pf on plant growth of *Solanum lycopersicum* L., varieties NS 528 and NS 585

1. NS 524 variety
A. Noninoculated control
B. Glomus fasciculatum (Thaxter) Gerdmann and Trappe emend. Walker & Koske
C. Gf+Ac
D. Gf+Pf
E. Gf+Ac+Pf

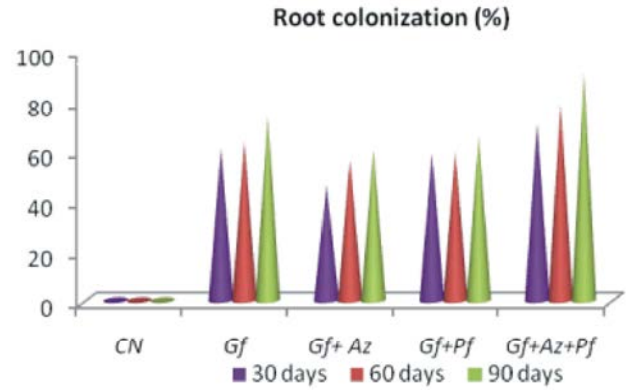
2. NS 585 variety
A. Noninoculated control
B. Glomus fasciculatum (Thaxter) Gerdmann and Trappe emend. Walker & Koske
C. Gf+Ac
D. Gf+Pf
E. Gf+Ac+Pf

significantly on fresh(13.06g), dry weight of shoot(1.22g), root length(15.24cm), fresh(1.07g), dry weight of root(0.28), number of leaves(45.33), stem diameter(3.10cm), root colonization(78.36%), phosphorus content(0.20%) in root was recorded highest inoculated with *Glomus fasciculatum, Azotobacter chrococcum* and *Pseudomonas fluorescens* than dual and single inoculation. There were no significant records in number of flower and fruits (Table 2).

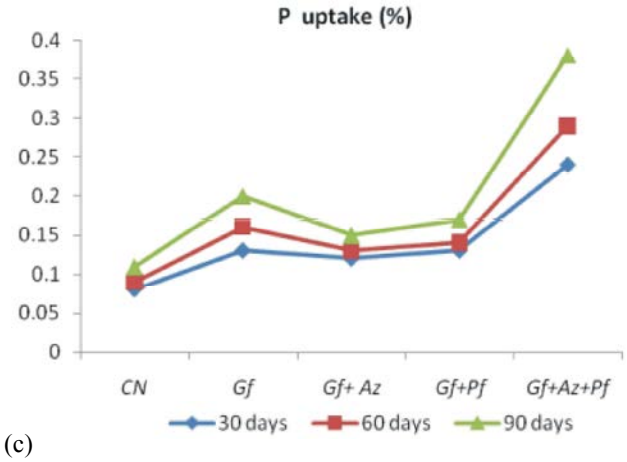
After 60 days the inoculation of *Glomus fasciculatum, Azotobacter chrococcum* and *Pseudomonas fluorescens* responded positively on shoot length(85.29 cm), fresh (92.54g) and dry weight of shoot (11.55g), root length(49.47cm), fresh (10.25g) and dry weight of root(2.56g) was recorded significantly. Root colonization (91.40%) and phosphorus content in root (0.20%) were observed in the inoculated *Glomus fasciculatum, Azotobacter chrococcum* and



(a)



(b)



(c)

Fig. 2: Showing effect of *Glomus fasciculatum, Azotobacter chrococcum* and *Pseudomonas fluorescens* on(a) dry weight of root (b) percent of root colonization and (c) Phosphorus uptake in *Solanum lycopersicum* L., (var .NS 524)

Pseudomonas fluorescens. The number of leaves(103.06), flowers(10.00) and fruits(5.66), stem diameter(3.68cm) and spore number(99.00) were highest inoculated with *Glomus fasciculatum, Azotobacter chrococcum* and *Pseudomonas fluorescens* compared to single and dual inoculation. After 90 days of plant growth, triple inoculation of *Glomus fasciculatum, Azotobacter chrococcum* and *Pseudomonas fluorescens* induced an increase in shoot length(89.15 cm), fresh (112.48g) and dry weight of root(19.77) than dual and single inoculations. The number of leaves (118.00), fruits(12.00), flowers(4.33), spore number(103.33) in 50g soil and root colonization(90.39%), phosphorus content in root(0.42%) and stem diameter(4.47cm). In comparison within dual

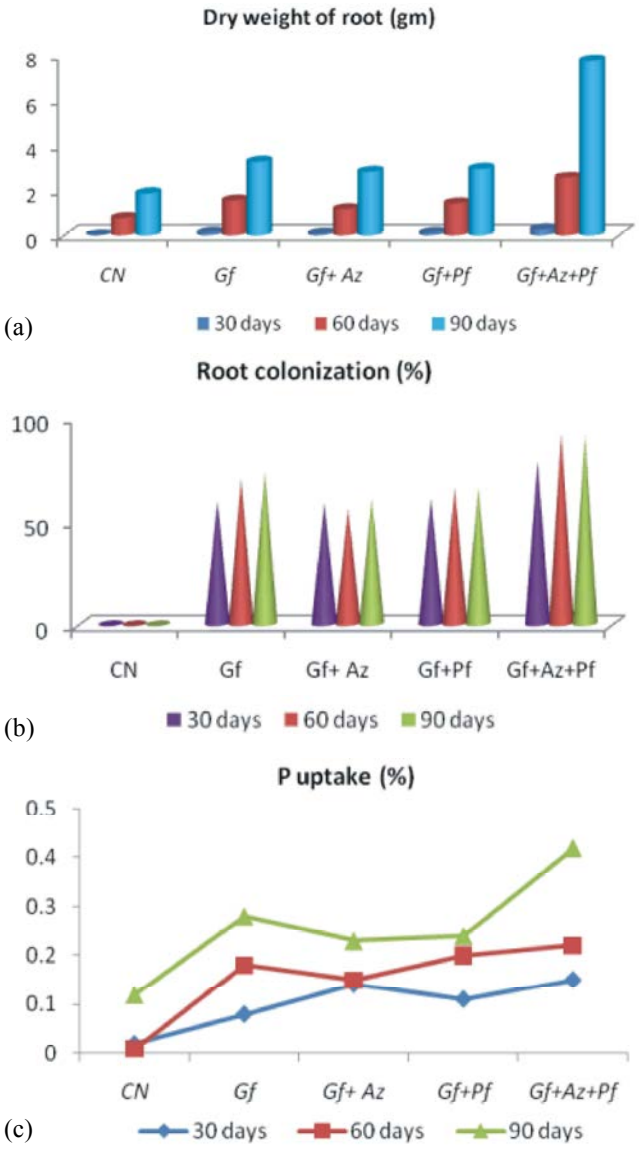


Fig. 3: Showing effect of *Glomus fasciculatum, Azotobacter chrococcum* and *Pseudomonas fluorescens* on (a) Dry weight of root (b) Percent of root colonization and (c) Phosphorus uptake in *Solanum lycopersicum* L., (Var .NS 585)

inoculation *Glomus fasciculatum* with *Pseudomonas fluorescens* resulted higher than *Glomus fasciculatum* and *Azotobacter chrococcum* and there is increase in single inoculated *Glomus fasciculatum* treatment, the dry weight of root, phosphorous uptake and root colonization (Fig. 3).

DISCUSSION

The results of the study [26] showed that seed priming with Plant Growth Promoting Rhizobacteria affected grain yield, plant height, number flowers, number of grains per ear significantly. Numerous other studies have shown a substantial increase in dry matter accumulation and seed yield following inoculation with PGPR [27, 28, 29]. [30] reported that in all their treatments, shoot and fruit potassium increased when PGPR and Arbuscular Mycorrhiza Fungi (AMF) were used together. The influence of PGPR on dry matter accumulation and chick pea (*C. arietinum* L.) yield under field conditions has been thoroughly studied [31]. Synergistic effects of plant growth-promoting

rhizobacteria andRhizobiumon nodulation and nitrogen fixation by pigeonpea (*Cajanus cajan*) were also observed [32]. The PGPR promote the growth of the plant and increase the root surface area or the general root architecture [33, 34]. An increase in chlorophyll content due to inoculation with AM fungi has been reported previously [35, 36]. The present study shows that co-inoculation of bacterial strains with AM fungi can enhance the plant growth significantly. The beneficial effect of co-inoculation on nutrient uptake could increase the volume of root and overall growth of the plant. Tomato plants also attributed to good uptake of phosphorous. Bacteria may also support the AMF symbiosis by increasing bio-available phosphate. In soil with low P bioavailability, free-living phosphate-solubilising bacteria may release phosphate ions from sparingly soluble inorganic and organic P compounds in soil [37]. While enhancement of mycorrhizal colonization resulting in improved nutrient (P) uptake by cereals under mixed cropping of 'cereals - non-legumes' or 'cereal - cereal' combinations like rice (*Oryza sativa* L.) Finger millet (*Eleusine coracana* L. Gaertn) [38] was attributed to higher root density per volume of soil favoring the spread of the symbiotic fungi [39]. The positive effect of PGPR and AMF bio-inoculants on wheat growth, observed in previous greenhouse experiments [40], was confirmed in the field. The bioinoculations had a significant effect on grain quality, for instance the phosphorus content that doubled in the bioinoculated plants. In our study, there were several cases in which AMF and PGPR coinoculations supported each other in terms of the improvement of plant growth and nutrient uptake, particularly phosphorus uptake, which also supports by [41,42]. For maximizing the better growth and vigour of *Eugenia bacterta* and *Terminalia bellerica* seedlings at nursery level, combined application of PSB, VAM and Rock phosphate is recommended to healthy timber seedlings [43].

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