

Original Research Article

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Effect of Plant Growth Promoting Bacteria on Seed Germination, Seedling Vigor and Growth *Lagenaria siceraria* (Molina) Standl

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ABSTRACT

This scrutiny was conducted to ascertain the effect of plant growth-promoting bacteria on seed germination, seedling vigor and growth and *Lagenaria siceraria* (Molina) Standl. was conducted during Kharif season of 2018-2019, 2019-20 at the University Farm, University College of Agriculture, Talwandi Sabo, Bathinda (Punjab). The experiment was laid out in RBD design with three replications. The crops were sown in poly-bags with a capacity of 10 kg which is filled with mixture formaldehyde sterilized coarse sandy loam soil and poly-bags are arranged according to the treatments and at a recommended spacing of crops. Different strains of bacteria, *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC8141, were used alone, mixer as a bio-priming agent with RDF 50% and 100% in the field conditions. A total of seven treatments are crafted included control (water-soaked) treatment. The treatment RDF 50% + *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC8141 affects the vine length, number of nodes on the main axis, number of leaves, leaf area, days to first fruit set, and also the number of primary branches per vine in bottle gourd crop significantly from other treatments. The highest germination percentage and seedling vigor were obtained from RDF 100% + *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC8141 treatment in bottle gourd crop as compared to that of the other treatments. The results of this study showed that and RDF 100% + *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC8141 treatment maximize the germination percentage and seedling vigor. The treatment RDF 50% + *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC8141 has a great potential to increase growth parameters bottle gourd crop.

Keywords

Serratia marcescens,
Bacillus subtilis,
PGPR and bottle
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Introduction

Bottle gourd [*Lagenaria siceraria* (Molina) Standl] is a vital cucurbitaceous crop grown for its fleshy fruits in tropical and subtropical regions (Desai *et al.*, 2016). It's probably

originated in Africa. Bottle gourd young fruits are edible, but the mature hollowed shells used as bottles, musical instruments, and floats for fishermen. Bottle gourd fruit crush is employed within the treatment of insanity, epilepsy, and other nervous diseases. Bottle

gourd plant is an annual herb and vigorous running vine. Stems of bottle gourd are succulent angular, the leaf is simple, sub-orbicular non-lobed or slightly lobed; flowers white, monoecious, and axillary; fruits are berry, varies in size, and shape, woody when mature. Bottle gourd area in India was 186 million hectares and production are 3052 '000' million tonnes during 2018-2019 (Agricultural statistics, 2019). The mechanisms by which PGPRs promote plant growth aren't fully understood. But, several mechanisms are suggested by which PGPR can enhanced stress resistance, symbiotic N₂ fixation according to Salantur *et al.*, (2006), solubilization of phosphate, and mineralization of organic phosphate or other nutrients (Cattelan *et al.*, 1999; Jeon *et al.*, 2003), increasing the availability or availability of primary nutrients to the host plant and antagonism against phytopathogenic microorganisms by the production of siderophores, synthesis of antibiotics, enzymes or fungicidal compounds and competition with detrimental microorganisms (Ashrafi *et al.*, 2011; Wu *et al.*, 2005; Lucy *et al.*, 2004; Ahmad *et al.*, 2005; Egamberdiyeva *et al.*, 2007). According to Dursun *et al.*, (2019) the seeds of tomato cultivars treated with three different concentrations (1, 3, and 5 gL⁻¹ and control i.e. un-inoculated) and two different bacterial fertilizers including *Azotobacter* spp. (1×10⁹ CFU), and a mixture of *Bacillus subtilis* and *Bacillus megatarium* (1×10⁹ UFC). The effects of these treatments were found significant in plant growth parameters. Bacterial fertilization increased yield and other parameters in all treatments of tomato. Kumar *et al.*, (2017) studied that the effect of *Enterobacter*, *M. arborescens* and *Serratia marcescens* on yield and nutrient uptake of wheat and suggested that consortium of two or three isolates significantly increased plant height (13.91%, 34.32%), straw yield (78.58%, 26.23%) and grain yield (79.83%, 24.05%) in pot and field

experiment respectively and uptake of nutrients such as N by 50.64% and P by 56.49% was also enhanced by wheat in field conditions was also observed. Singh *et al.*, (2017) suggested that *Pseudomonas* (RS1) and *Bacillus* (R7) most dominant in the rhizosphere of bitter melon and *Bacillus* (R7) may be used as a biofertilizer for bitter melon growth. However, scanty information is out there regarding the utilization of bio-priming in several crops and it must be investigated. Keeping in sight the above facts this investigation is going to be undertaken to review the effect of plant growth-promoting bacteria on seed germination, seedling vigor and growth of *Lagenaria siceraria* (Molina) Standl. with the objectives to evaluate the effect of plant growth-promoting bacteria on seed germination, seedling vigor and growth of bottle gourd.

Materials and Methods

The present investigation entitled "Effect of plant growth-promoting bacteria on seed germination, seedling vigour and growth of *Lagenaria siceraria* (Molina) Standl." was conducted during Kharif season of 2019 and 2020 at the University College of Agriculture farm, Talwandi Sabo, Bathinda (Punjab) and experiment was laid out by Randomized Block Design with three replications. Three standard PGPR strains these included *Serratia marcescens* MTCC 10241 and *Bacillus subtilis* MTCC7611; MTCC814 were obtained from Microbial Type Culture Collection (MTCC) Chandigarh, India.

Sub culturing of bacterial culture

The bacterial culture was sub cultured by growing in nutrient broth and centrifuged at 120 rpm for 24 hrs. Spectrophotometer at "600" nm light was used to check the growth of bacteria optical density.

Sterilization and inoculation of seeds

The seeds of bottle gourd was surface sterilized with NaOCl for 2 minutes, and after that wash thoroughly with sterile distilled water for three times. Then surface sterilizes seeds was soaked into bacterial suspension at a particular concentration and they were allowed for overnight. The seeds were soaked in sterile distilled water was used as control.

Preparation of pots and treatment details

The soil was sterilized with formalin @ g/m³ before two weeks of transplanting. Poly-bags with a capacity of 10 kg are filled with mixture formalin sterilized coarse sandy loam soil and treated seeds of bottle gourd variety Punjab Bahar were sown. The poly-bags were arranged according to the recommended spacing in the open field. Total treatments are seven and these were T₁: Control (Water soaked), T₂: Fertilizer 100% + *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC8141, T₃: Fertilizer 50% + *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC8141, T₄: *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC8141, T₅: *Serratia marcescens* MTCC 10241, T₆: *Bacillus subtilis* MTCC8141, T₇: *Bacillus subtilis* MTCC761.

Details of observations recorded

Seed germination percentage was analysed by using the formula: Seed Germination % = Germinated seeds / Total Seed Sown × 100, Seed vigour was measured by using formula: Seedling length × % germination, Vine length (cm.), Number of primary branches per vine (No.), Number of nodes on the main axis (No.), Number of leaves (No.), Leaf area (cm²) and Days to first fruit set (No.) are parameters observed during field trail in the

2019 and 2020 kharif season.

Statistical analysis

All data were analysed for different characters with the help of OPSTAT (Sheoran *et al.*, 1998). The critical difference at 5% level of implication was calculated to equate the mean different treatments.

Results and Discussion

The data in table 1 shows that treatment T₂ (RDF 100 % + *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) had the significant results in seed germination percentage (89.27 %) in 2019 and (88.85 %) in 2020 years of experimentation. And it was followed by T₃ (RDF 50 % + *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814), the seed germination percentage was 86.92 % in 2019 and 85.61 % in the 2020. Seedling vigor significantly higher in 2019 (2050.89) and in 2020 (2048.17) by T₂ (RDF 100 % + *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) treatment and it was followed by T₃ (RDF 50 % + *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) treatment in both 2019 (1932.76) and in 2020 (2025.43) years.

The table 2 revealed that T₃ (RDF 50 % + *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) treatment shows the significant result of vein length in both 2019 (253.51 cm) and 2020 (251.51 cm) years and followed by treatment T₂ (RDF 100 % + *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) in both years. The observation was 248.76 cm vein length in 2019 and 246.61 cm in the 2020. Treatment T₃ (RDF 50 % + *Serratia*

marcescens MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) shows the significant result in the number of primary branches per plant of bottle gourd crop in both 2019 (4.73) and 2020 (4.47) years and it was followed by treatment T₂ (RDF 100 % + *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) in 2019 the number of primary branches per plant was 4.24 and 4.04 number of primary branches per plant in the year 2020.

The table 3 shows that the number of nodes on the main axis significantly maximum in treatment T₃(RDF 50 % + *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) in 2019 (21.33) and the 2020 year observation was same as in 2019 and followed by T₂ treatment with observation 19.46. Treatment T₂(RDF 100 % + *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) and T₄ (*Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) was at par with each other and followed the T₃ treatment in the 2020 year. Maximum number of leaves per plant 32.38 were found in the treatment T₃ (RDF 50% +*Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) and followed by treatment T₂(RDF 100% +*Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) with observation 30.13 in the 2019 year. A maximum number of leaves per plant 30.90 are reported in the 2020 year in by T₃ (RDF 50% +*Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) treatment and followed by T₂ (RDF 100% +*Serratia marcescens* MTCC10241 + *Bacillus subtilis*

MTCC7611 + *Bacillus subtilis* MTCC814) treatment with 28.43 number of leaves per plant and it was at par with 27.30 number of leaves per plant by T₄ *Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) treatment.

The table 4 shows that maximum leaf area 187.99 cm² were found in the treatment T₃ (RDF 50% +*Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) and followed by treatment T₂ (RDF 100% +*Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) with observation of leaf area 182.60 cm² in 2019 year. Maximum leaf area 187.25 cm² are reported in 2020 year in by T₃ (RDF 50% +*Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) treatment and followed by T₂ (RDF 100% +*Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) treatment with 182. 45 cm² leaf area. The days to first fruit set (48.32) were found significantly reduced in the treatment T₃ (RDF 50% +*Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) and followed by treatment T₂(RDF 100% +*Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) with observation 50.66 days to first fruit set in the 2019 year. Day to first fruit set (37.90 days) are reported in the 2020 year in by T₃ (RDF 50% +*Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) treatment and it was at par with T₂ (RDF 100% +*Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) treatment with 49 days to first fruit set.

Table.1 Effect of plant growth-promoting bacteria on seed germination percentage and seedling vigor of *Lagenaria siceraria* (Molina) Standl

Parameter	Seed germination percentage			Seedling vigor			
	Treatments	2019	2020	Pooled	2019	2020	Pooled
T ₁		63.00	61.5	62.25	1006.48	1005.63	1006.06
T ₂		89.09	88.61	88.85	2050.89	2048.17	2049.53
T ₃		86.92	85.70	86.31	1932.76	2025.43	1979.10
T ₄		71.01	70.73	70.87	1414.79	1446.20	1430.50
T ₅		61.19	60.48	60.83	1047.33	1036.00	1041.67
T ₆		58.51	63.43	60.97	1028.11	1031.90	1030.01
T ₇		63.67	64.41	64.04	1281.53	1180.57	1231.05
CD (0.05)		1.06	0.92	1.54	0.76	4.31	3.41

Table.2 Effect of plant growth-promoting bacteria on vine length and number of primary branches per vein of *Lagenaria siceraria* (Molina) Standl

Parameter	Vein length (cm)			Number of primary branches per vein (No.)			
	Treatments	2019	2020	Pooled	2019	2020	Pooled
T ₁		166.33	165.05	165.69	3.24	2.30	2.77
T ₂		248.76	246.61	247.69	4.24	4.04	4.14
T ₃		253.51	251.60	252.56	4.73	4.47	4.60
T ₄		240.69	243.85	242.27	3.66	3.92	3.79
T ₅		180.96	179.09	180.02	3.27	3.27	3.27
T ₆		210.58	194.01	192.31	3.35	3.58	3.47
T ₇		190.61	207.16	208.87	3.44	3.37	3.40
CD (0.05)		0.62	4.09	2.45	0.07	0.10	0.26

Table.3 Effect of plant growth-promoting bacteria on the number of nodes on the main axis and number of leaves of *Lagenaria siceraria* (Molina) Standl

Parameter	Number of nodes on the main axis(No.)			Number of leaves (No.)			
	Treatments	2019	2020	Pooled	2019	2020	Pooled
T ₁		16.40	15.40	15.90	23.14	22.54	22.84
T ₂		19.46	18.33	18.90	30.13	28.43	29.28
T ₃		21.33	21.33	21.33	32.38	30.90	31.64
T ₄		18.33	18.00	18.16	27.13	27.30	27.21
T ₅		16.00	16.00	16.00	24.43	23.84	24.14
T ₆		15.33	16.66	16.00	25.31	24.97	25.14
T ₇		17.66	17.66	17.66	25.90	25.52	25.71
CD (0.05)		0.82	0.87	0.72	0.40	1.30	0.71

Table.4 Effect of plant growth-promoting bacteria on leaf area and days to first fruit set of *Lagenaria siceraria* (Molina) Standl

Parameter	Leaf area (cm ²)			Days to first fruit set (days)			
	Treatments	2019	2020	Pooled	2019	2020	Pooled
T ₁		166.40	162.50	164.45	58.37	57.48	57.92
T ₂		182.60	182.45	182.52	50.66	49.00	49.83
T ₃		187.99	187.25	187.62	48.32	47.90	48.11
T ₄		177.87	177.35	177.61	54.06	52.33	53.19
T ₅		169.90	169.04	169.47	57.23	56.46	56.84
T ₆		173.54	173.13	173.33	58.60	58.20	58.40
T ₇		174.78	173.70	174.24	55.15	53.66	54.41
CD (0.05)		2.72	1.89	1.61	0.51	1.40	0.72

Seed germination percentage and seedling vigor

Germination percent and seedling length are the major factors for deciding the seedling vigor. In the present study, this character showed significant variation among all treatments which might be due to PGPR (*Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) activity. Similar results were also reported by (Prathibha and Siddalingeshwara, 2013) studied the effect on seed treatment with PGPRs such as *Pseudomonas fluorescens* and *Bacillus subtilis* significantly increased seed germination, vigor index, and nutritional quality.

Vein length and the number of primary branches per plant

These are the characters of growth which should be enhanced to get optimum yield from the crop. So, mixture of all the bacteria (*Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) with 50% recommended dose of fertilizer in this experiment significantly increase the vein length and the number of primary branches per plant of bottle gourd. Correspondingly Kumar *et al.*,

(2015) studied that seed coating with *Bacillus subtilis* OTPB1 and *Trichoderma harzianum* OTPB3 of brinjal, beans, bitter gourd, bottle gourd, cabbage, chili, carrot, cauliflower, pumpkin, ridged gourd), fruit crop (papaya in plastic trays in a glasshouse), tuber crops (potato), ginger and turmeric, a significant increase in growth parameters under greenhouse conditions and growth and yield parameters under field conditions were recorded.

The number of nodes on the main axis and number of leaves per plant

The results revealed that the T₃ (RDF 50% +*Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) treatment shows significantly increase the number of nodes on the main axis in bottle gourd crop during both the years. On the contrary Kidoglu *et al.*, (2008) studied that *Pseudomonas putida*, *Enterobacter cloacae*, *Serratia marcescens*, *Pseudomonas fluorescens*, *Bacillus* spp., *Pseudomonas putida*) significantly increases in growth of cucumber, tomato, and pepper. The data with regards to the number of leaves produced by each plant was maximum observed in the treatment T₃ (RDF 50% +*Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus*

subtilis MTCC814) in both years. Similarly, Yıldırım *et al.*, (2015), applied *Bacillus pumilis* and *Alcaligenes piechaudii* strains as seed and/or drench treatments and found increased number of leaves in cucumber.

Leaf area and the days to first fruit set

In the pooled data with regards to leaf area produced by each plant was maximum observed in the T₃(RDF 50% +*Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) treatment in bottle gourd crop in both years. Equally Mia (2010) evaluated the effect of PGPR on banana plantlets under nitrogen-free hydroponic condition and found an increase in growth attributes such as root hair, leaf area, chlorophyll content and total biomass. In pooled data T₃(RDF 50% + *Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) treatment was significantly superior for a reduced number of days to first fruit set in bottle gourd crop during both years of investigation. Similarly Karakurt *et al.*, (2011) also found that *B. subtilis* OSU-142, *B. megaterium* M-3, *B. cepaciai* OSU-7, and *P. putida* BA-8 have great potential to increase fruit set, plant growth, and fruit quality. Present study support that use of PGPRs may enhance the seed germination, seedling vigor, and growth of bottle gourd crop under field condition. The effective PGPR strain may be recommended to farmer which may reduce the use of chemical fertilizers in cucurbit crops.

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