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Effect of plant growth promoting bacteria on seed germination, seedling vigor and growth of *Cucumis sativus* L.

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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 of *Cucumis sativus* L. 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 significantly affects the seed germination percentage, seedling vigor, vine length, number of nodes on the main axis, number of leaves, leaf area and days to first fruit set in cucumber crop from other treatments. The highest number of primary branches per vine in cucumber obtained from RDF 100% + *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC8141 treatment as compared to that of the other treatments. It was concluded from study that PGPR with 50% RDF significantly improved the seed germination percentage, seedling vigor and growth parameters of cucumber crop.

Keywords: *Serratia marcescens, Bacillus subtilis*, cucumber and bottle gourd

Introduction
Cucumber (*Cucumis sativus* L.) is indigenous to the tropical and subtropical. Cucumber wild species Centre of origin is Africa and cultivated species (*C. s.* var. *sativus*) may have originated in Africa, China, and India or within the Nearest East. It required a warm climate for rapid and satisfactory production. Cucumber is the hottest vegetable cops used everywhere on the planet in several ways. The fruit is employed as table salad, eaten raw, with the standard salad seasonings, and is pickled in large quantities. The cucumber plant is annual monoecious with tailing stems, roots are extensive and largely superficial, leaves alternate with 3-7 lobed, flowers monoecious or unisexual, and fruits are globose to cylindrical berry. In India, the total, area under vegetable crops is 10100 million hectares and production is 185883 '000' million tonnes. Cucumber area is 109 million hectares and production is 1696 '000' million tonnes in 2018-19 (Agricultural statistics, 2019). Excessive chemical fertilizers and pesticide usages in crop production persuade the risks of both human health and the environment. One of the opposites most vital effective factors in increasing plant yield is seed inoculation or priming with the plant growth-promoting rhizobacteria (PGPR) reported by (Ashraf *et al*. 2011) [3]. Also, plant growth-promoting rhizobacteria (PGPR) are a gaggle of bacteria that actively colonize plant roots and increased plant growth and yield (Rao, *et al*. 1999; Wu, *et al*. 2005; Heidari, *et al*. 2011) [10, 12, 7]. According to Alaaeldin *et al*. (2018) [2] the use of PGPR was environmentally sound way of decreasing chemical fertilizers and increasing crop yields. Avinash *et al*. (2017) [4] revealed that *Bacillus amyloliquefaciens* MIC6 and *Pseudomonas aeruginosa* MTCC2581 showed maximum germination and yield of Cucumber manages *Fusarium* wilt both in protected and open conditions. Batool and Altaf (2017) [5] also concluded that the effect of fertilizer and plant growth-promoting rhizobacteria on growth, the yield of chili (*Capsicum frutescens* L.) ($N_{100\%}$ $P_{75\%}$ $K_{100\%}$+PGPR) showed better results compare to other treatments. 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 *Cucumis sativus* L.

with the objective to evaluate the effect of plant growth-promoting bacteria on seed germination, seedling vigor and growth of cucumber.

Materials and Methods

The present investigation entitled "Effect of plant growth-promoting bacteria on seed germination, seedling vigour and growth of *Cucumis sativus* L." was conducted during Kharif season of 2018-2019 and 2019-20 at the University Farm, University College of Agriculture, Talwandi Sabo, Bathinda (Punjab) and experiment was laid out by Randomized Block Design with three replications. Three standard PGPR strains were obtained from Microbial Type Culture Collection (MTCC) Chandigarh, India and these included *Serratia marcescens* MTCC 10241 and *Bacillus subtilis* MTCC7611; MTCC814. The bacterial culture was sub cultured by growing in nutrient broth and centrifuged at 120 rpm for 24 hrs. The purity of the bacterial culture was checked by staining methods and store at 4 ºC in slant culture for further studies. To check the growth of bacteria optical density was measured under spectrophotometer in "600" nm light. The seeds of Cucumber was surface sterilized with 0.4% sodium hypochlorite (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 (Approx. 100 cells of *S. Marcescens* /seed and Approx. 100 cells of *B. Subtilis* /seed) separately, and they were allowed for overnight. The seeds were soaked in sterile distilled water was used as control. The soil was sterilized with formaldehyde @ g/m^3 (Adjustment of a dose is according to temperature i.e. 16-20ºC @ 8g/m^3 and at the interval of 5 ºC below 20ºC the dose will be doubled) before 2 to 3 weeks of transplanting. Poly-bags with a capacity of 10 kg are filled with mixture formaldehyde sterilized coarse sandy loam soil and treated seeds of cucumber variety Punjab Naveen were sown. The poly-bags were arranged according to the treatments. Total treatments are seven i.e. T_1: Control (Water soaked), T2: Fertilizer 100%+ *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC8141, T_3: Fertilizer 50% + *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC8141, T_4: *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC8141, T_5: *Serratia marcescens* MTCC 10241, T_6: *Bacillus subtilis* MTCC8141, T_7: *Bacillus subtilis* MTCC761. Seed germination percentage was analyzed 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 (cm^2) and Days to first fruit set (No.) are parameters observed during research trail in both years. All data from the experimental field were analysed for different characters with the help of OPSTAT (Statistical Software Package for Agricultural Research Workers) (Sheoran *et al*., 1998) [11]. The critical difference at 5% level of implication was calculated to equate the mean different treatments.

Results and Discussion

An examination of the data indicated (Table 1) that all the concentrations of plant growth-promoting bacteria increase the seed germination percentage significantly as compared to C_1 (control). T_3 (RDF 50% + *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) shows significant result in seed germination percentage of cucumber in both 2019 (84.78%) and 2020 (83.27%) years. It was followed by T_4 (*Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) in both 2019 (82.09%) and 2020 (81.61%) years of experimentation. Seedling vigor 1321.33 in 2019 and 1318.33 in 2020 year in cucumber crop significantly also recorded higher in T_3 (RDF 50% + *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) and followed by T_4 (*Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) treatment in both 2019 (1281.22) and the 2020 (1278.45) regarding 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 activity. Similar results were also reported by Yıldırım *et al.* (2015) [13] that the effects of bacterial applications depend on the crop species, the results showed that *Bacillus pumilis* and *Alcaligenes piechaudii* have a great potential in increase cucumber seedling growth and quality.

Table 1: Effect of plant growth-promoting bacteria on seed germination percentage and seedling vigor of *Cucumis sativus* L.

Parameter	Seed germination percentage			Seedling vigor		
Treatments	**2019**	**2020**	**Pooled**	**2019**	**2020**	**Pooled**
T_1	68.98	67.23	68.10	870.82	869.72	870.27
T_2	81.57	79.27	80.42	1215.72	1213.52	1214.62
T_3	84.78	83.27	84.02	1321.33	1318.33	1319.83
T_4	82.27	81.61	81.94	1281.22	1278.45	1279.83
T_5	59.09	59.26	59.18	846.49	844.49	845.49
T_6	65.34	64.48	64.91	914.83	913.57	914.20
T_7	71.42	69.48	70.45	1071.30	1068.10	1069.70
CD (0.05)	0.58	0.17	0.61	0.25	0.24	1.42

In the table 2 shows that the treatment T_3 (RDF 50% + *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) shows the significant result of vein length in both 2019 (89.27 cm) and 2020 (87.27 cm) years and followed by T_4 (*Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) in 2019 vein length was 83.50 cm and in 2020 vein length was 82.78 cm. Treatment T_2 (RDF 100% + *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) significantly increased the number of primary branches per plant in 2019 (3.94) and the number of primary branches per plant was non-significant in the 2020. Similar results were reported by Batool and Altaf (2017) [5] that the effect of fertilizer and plant growth-promoting rhizobacteria on growth, the yield of chili (*Capsicum frutescens* L.) ($N_{100\%}$ $P_{75\%}$ $K_{100\%}$ + PGPR) showed better results compare to other treatments.

Table 2: Effect of plant growth-promoting bacteria on vine length and number of primary branches per vein of *Cucumis sativus* L.

Parameter	Vein length (cm)			Number of primary branches per vein		
Treatments	**2019**	**2020**	**Pooled**	**2019**	**2020**	**Pooled**
T_1	64.36	63.33	63.84	1.93	1.87	1.90
T_2	78.40	76.32	77.36	3.94	3.92	3.93
T_3	89.49	87.27	88.38	3.86	3.84	3.85
T_4	83.50	82.78	83.14	3.44	3.43	3.43
T_5	59.36	60.11	59.74	1.74	1.62	1.68
T_6	62.71	64.46	63.58	2.23	2.14	2.18
T_7	74.15	72.26	73.21	2.73	2.53	2.63
CD (0.05)	0.44	0.69	1.04	0.06	0.04	0.05

Data in the table 3 showed marked improvement in the number of nodes on the main axis with treatment T_3 (RDF 50% + *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) in both 2019 (24.66) and 2020 (24.33) years in cucumber crop and it was followed by T_2 (RDF 100% + *Serratia marcescens* MTCC 10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) in both 2019 (22.26) and 2020 (22.33) years. The maximum number of leaves per plant 26 were found in the treatment T_3 (RDF 50% + *Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) and followed by treatment T_2 (RDF 100% + *Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) with observation 24 and it is at par with T_4 (*Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) with 23 number of leaves per plant in the 2019 year. The maximum number of leaves per plant 25.33 are reported in the 2020 year in cucumber crop by T_3 (RDF 50% + *Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) treatment and followed by T_2 (RDF 100% + *Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) treatment with 23.66 number of leaves per plant and it was at par with treatment T_4 (*Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) with a record of 22.33 number of leaves per plant. In the same way Ibiene *et al*. (2012) [8] used the PGPR as bio-fertilizer in increased growth parameters (plant height, stem width, root length, internode length) of tomato transplant compared to control.

Table 3: Effect of plant growth-promoting bacteria on node number for the number of nodes on the main axis and Number of leaves of *Cucumis sativus* L.

Parameter	Number of nodes on the main axis			Number of leaves		
Treatments	**2019**	**2020**	**Pooled**	**2019**	**2020**	**Pooled**
T_1	15.00	14.33	13.83	13.00	12.33	13.83
T_2	22.66	22.33	22.50	24.00	23.66	22.50
T_3	24.66	24.33	24.50	26.00	25.33	24.50
T_4	20.66	20.33	20.50	23.00	22.33	20.50
T_5	17.33	17.00	17.16	13.33	12.33	17.16
T_6	18.33	17.66	18.00	15.66	14.66	18.00
T_7	21.00	20.33	20.66	18.66	18.33	20.66
CD (0.05)	1.28	0.98	0.75	1.48	1.01	0.75

The table 4 shows the result that maximum leaf area 16.27 cm^2 were found in the treatment T_2 (RDF 100% + *Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) and followed by treatment T_3 (RDF 50% + *Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) with leaf area 15.23 (cm^2) in 2019 year. Maximum leaf area 15.26 (cm^2) are reported in 2020 year by T_2 (RDF 100% + *Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) treatment and followed by T_3 (RDF 50% + *Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) treatment with 14.37 cm^2 leaf area. During 2019 Kharif season the treatment T_3 (RDF 50% + *Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) were reduced the days to first fruit set up to 47.33 days and it was at par with treatment T_4 (*Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) with observation 47.66 days and it was followed by T_2 (RDF 100% *Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) with 49.33 days to first fruit set in the 2019 year. Reduced number of days to first fruit set 46.33 are reported in the 2020 year by T_3 (RDF 50% + *Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) treatment and it was at par with T_4 (*Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) treatment with 46.66 days to fruit set and it was followed by the treatment T_2 (RDF 100% + *Serratia marcescens* MTCC10241 + *Bacillus subtilis* MTCC7611 + *Bacillus subtilis* MTCC814) with a record of 48.66 days to first fruit set. Likewise Ning *et al*. (1997) [9] and Grelalorenzo *et al*. (1988) [6] found that applying N-Fixers increased fruit set and yield of pumpkins and tomato, respectively. It was concluded from study that PGPR with 50% RDF significantly improved the seed germination percentage, seedling vigor and growth parameters of cucumber crop. The effective PGPR strain may be recommended to farmer which help to reduce their dependence on inorganic chemicals. Such strategy may also help in the vegetable production in organic farming.

Table 4: Effect of plant growth-promoting bacteria on leaf area and days to first fruit set of *Cucumis sativus* L.

Parameter	Leaf area (cm2)			Days to first fruit set (days)		
Treatments	**2019**	**2020**	**Pooled**	**2019**	**2020**	**Pooled**
T_1	11.33	11.22	11.28	52.33	51.66	52.00
T_2	16.27	15.26	15.77	49.33	48.66	49.00
T_3	15.23	14.37	14.80	47.33	46.33	46.83
T_4	12.45	12.87	12.66	47.66	46.66	47.16
T_5	11.42	11.34	11.38	54.33	51.66	53.00
T_6	11.71	11.53	11.62	51.66	50.33	51.00
T_7	12.12	12.43	12.27	53.33	51.66	52.50
CD (0.05)	0.39	0.32	0.41	1.08	1.22	0.82

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