Effect of different microbial inoculants on morphological and fruit characteristics of strawberry (*Fragaria* × *ananassa*) cv. winter dawn

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Strawberry (Fragaria × ananassa) a hybrid between two American species Fragaria virginiana × Fragaria chiloensis is a perennial plant belonging to the family Rosaceae. It is one of the most, delicious, attractive and highly perishable fruit. It contains approximately 89.9% water and good source of vitamin C, phenols, flavonoids, and other nutrients like potassium and calcium (Roussos et al. 2022). In India, the area under the strawberry cultivation area is around 2000 ha (Anonymous 2021a). Strawberry requires the higher amount of nutrients for production of a better crop (Osvalde et al. 2023). Farmers rely on chemical fertilizers and insecticides to improve crop yield. Sadly, several researchers have found lingering build-up of hazardous compounds in fruits. Biofertilizers are a good replacement for chemical or synthetic fertilizers and are becoming increasingly significant in agriculture. Microbial inoculants such as arbuscular mycorrhiza fungi (AMF) and plant growth promoting bacteria (PGPB) are used as biofertilizers to improve the availability and uptake of soil nutrients (Alori et al. 2017). Microbes likes Pseudomonas, Bacillus, Rhizobium and Burkholderia can solubilize inorganic phosphorus and make it in an available form to the plants, Acinetobacter, Alcaligenes, Arthrobacter, Bacillus, Burkholderia, Pseudomonas, and Serratia are the most common plant growth-promoting bacteria (Mahanty et al. 2016). Meager studies have been conducted to experiment the impression of biofertilizers on strawberry yield, fruit quality, and disease occurrence. The main aim of this research is to examine the effects of different microbial inoculants or rhizobacteria like Burkholderia spp. and Stenotrophomonas spp. etc. on plant vigour, fruit yield of strawberry plant.

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An experiment was conducted during 2022-23 at Regional Research Station (Punjab Agricultural University, Ludhiana), Bathinda, Punjab. The strawberry seedlings were transplanted on the raised bed at 25 cm × 25 cm spacing having 28 plants/plot under randomised block design (RBD) with four replications. Strawberry plants were grown and maintained by using the cultural practices as described in Anonymous (2021b). The strawberry roots were inoculated by different microbial @10 ml in 10 litre water with following treatments, T₁, Control; T₂, Burkholderia seminalis; T₃, Stenotrophomonas maltophilia; T₄, Vesicular arbuscular mychorrhizae (VAM); T₅, PAU consortium; T₆, ${\it Burkholderia\ seminalis\ +\ Stenotrophomonas\ maltophilia;}$ T_7 , Burkholderia seminalis + VAM; T_8 , Stenotrophomonas maltophilia + VAM; T₉, Burkholderia seminalis + PAU consortium; T₁₀, Stenotrophomonas maltophilia + PAU consortium; T₁₁, Burkholderia seminalis + VAM + Stenotrophomonas maltophilia + PAU consortium.

The plant observations, viz. plant height (cm) of strawberry was determined from the base of the crown to the top of the primary leaves by using a measuring scale. The plant spread (cm) was calculated by measuring the canopy of the plant with a scale in east-west and north-south directions and taking the average of both directions as a plant spread. The leaf area (cm²) was estimated by leaf area meter 211 (SYSTRONICS). Crown diameter (cm) was taken with the help of digital Vernier caliper. The number of leaves was calculated after 120 days of transplanting; the average leaf number/plant was recorded from tagged plants/replication in each treatment and indicated as mean leaf number/plant. The root observations, viz. the fresh weight (g) of strawberry roots was estimated during first fortnight of April. After uprooting the strawberry plant, roots were washed properly and air dried to remove excess moisture. The root weight was measured with digital balance and average was estimated as the root fresh. After taking fresh root weight, roots were kept in oven at 70°C for drying till the weight was constant to estimate the dry weight of root. The root volume of strawberry was calculated by water displacement method and average was worked out in 'cc'. The root length (cm) of strawberry plants was measured with the help of a scale and average value was worked out. The fruit observations i.e. the fruit length (cm) was measured from the calyx end to the pointy end or apex of the fruit using verniercaliper's. Berry diameter (cm) was measured using verniercaliper's, primarily from the berries' shoulders. The total number of fruits/plant was counted from tagged plants starting from end December to end-March. The fresh fruit/berries weight (g) was taken from tagged plants and weighed by using an electronic weighing balance. Cumulative fruit yield/plant (g/plant) was calculated by adding the marketable yield obtained from all the harvesting dates from selected plants and values were averaged to record the data.

Statistical analysis: The data was analysed by ANOVA by using R studio and mean values were compared using the LSD test at 0.05% of probability.

Plant morphological characteristics: The plant inoculation with different microbial inoculants showed significant enhancement in plant height, plant spread, crown diameter, leaf area and number of leaves/plant of strawberry (Table 1). In winter dawn cultivar, the plants inoculated with Burkholderia seminalis + Stenotrophomonas maltophilia + VAM + PAU consortium attained maximal plant height (26.67 cm), plant spread (32.75 cm), leaf area (79.19 cm²), number of strawberry leaves (22) and crown diameter (2.17 cm) whereas in control (untreated plant) results showed minimum plant height (20.62 cm), plant spread (26.80 cm), leaf area (53.57 cm²), number of strawberry leaves (19.12) and crown diameter (1.45 cm). Similar results were reported by Andrade et al. (2019). The strawberry plant inoculated by the combination of Azopirillium (Ab-V5) + Burkholderia (CCMA 0056) + Enterobacter (CCMA 1285) showed the increase in plant height (23.6 cm) as compared to noninoculation (15.30 cm). Reddy et al. (2020) concluded that the inoculation with microbial inoculants like Azotobacter

or *Azospirillum* along with the addition of 100% N fertilizer enhanced the strawberry plant spread by fixing and quick release of nitrogen to the plant. Negi *et al.* (2021), the plant height, spread, number of leaves/plant and leaf area of strawberry were improved by the application of FYM + vermicompost + *Pseudomonas* + *Azotobacter* because both inoculants improve the IAA synthesis which further promotes the vegetative growth and photosynthetic activity of plant.

Root characteristics: Microbial inoculants played positive role in increasing the root growth of strawberry (Table 2). The plant inoculated with a combination of Burkholderia seminalis + Stenotrophomonas maltophilia

Table 2 Effect of different microbial inoculants on root characteristics of strawberry

Treatment	Root fresh	Dry root	Root	Root
	weight	weight	length	volume
	(g)	(g)	(cm)	(cc)
Control	9.93 ^e	1.72 ^f	16.39 ^c	4.95 ^d
Burkholderia seminalis (BS)	11.50 ^{cde}	2.80 ^{cde}	17.70 ^c	6.25 ^{bc}
Stenotrophomonas maltophilia (SM)	11.15 ^{de}	2.68 ^{de}	17.62 ^c	6.13 ^{bc}
VAM	11.10 ^{de}	2.52 ^e	17.30 ^c	6.05 ^{bc}
PAU consortium	10.79 ^e	2.47 ^e	16.83 ^c	5.13 ^c
BS + SM	12.89 ^{bc}	3.43 ^{bc}	21.15 ^{ab}	6.70^{b}
BS + VAM	13.83 ^{ab}	3.58 ^b	21.49a	7.18 ^{ab}
SM + VAM	12.89 ^{bc}	3.47 ^{bc}	21.25 ^a	6.95 ^b
BS + PAU consortium	12.84 ^{bc}	3.21 ^{bcd}	18.75 ^{bc}	6.68 ^b
SM + PAU consortium	12.66 ^{bcd}	3.12^{bcde}	18.50 ^c	6.33bc
BS + SM + VAM + PAU consortium	14.91 ^a	4.40 ^a	21.57 ^a	8.40 ^a
LSD (<i>P</i> ≤0.05%)	1.67	0.69	2.42	1.31

Treatment details are given under Materials and Methods.

Table 1 Effect of different microbial inoculants on morphological characteristics of strawberry plant

Treatment	Plant height (cm)	Plant spread (cm)	Crown diameter (cm)	Number of leaves	Leaf area (cm ²)
Control	20.62 ^d	26.80e	1.45 ^c	19.12 ^d	53.57 ^e
Burkholderia seminalis (BS)	25.05 ^b	30.35 ^{bc}	1.85 ^{ab}	20.92 ^{bc}	70.92 ^{bc}
Stenotrophomonas maltophilia (SM)	25.02 ^b	30.35 ^{bc}	1.85 ^{ab}	20.73 ^{bc}	70.66 ^{bcd}
VAM	24.92 ^b	29.57 ^{cd}	1.80 ^{bc}	20.62bc	68.65 ^{cd}
PAU consortium	23.40°	27.95 ^{de}	1.62 ^{bc}	20.35 ^c	68.14 ^d
BS + SM	25.87 ^{ab}	31.27 ^{abc}	1.90 ^{ab}	21.02 ^{abc}	76.56 ^a
BS + VAM	26.47 ^a	31.90 ^{ab}	1.95 ^{ab}	21.45 ^{ab}	78.15 ^a
SM + VAM	25.90 ^{ab}	31.30 ^{abc}	1.92 ^{ab}	21.10 ^{abc}	76.67 ^a
BS + PAU consortium	25.67 ^{ab}	31.10 ^{abc}	1.90 ^{ab}	20.96bc	72.10 ^b
SM PAU consortium	25.15 ^b	30.75 ^{bc}	1.87 ^{ab}	20.90 ^{bc}	70.97 ^{bc}
BS + SM + VAM + PAU consortium	26.67 ^a	32.75 ^a	2.17 ^a	22.00 ^a	79.19 ^a
LSD (<i>P</i> ≤0.05%)	1.23	1.73	0.36	1.03	2.77

Treatment details are given under Materials and Methods.

Cumulative yield Treatment Fruit diameter Fruit length Fruit weight Number of fruits (g/plant) (cm) (cm) (g) Control 2.38^{b} 3.45^{d} 16.96^d 11.45^c 194.15^d 2.70ab 3.65abcd 18.06bcd 13.43ab 242.11bc Burkholderia seminalis (BS) 17.97bcd Stenotrophomonas maltophilia (SM) 2.60ab 3.58bcd 13.38ab 240.11bc 2.40^{b} 3.55bcd 17.52^{cd} 220.86cd VAM 12.61bc 3.53^{cd} 200.52^{d} PAU consortium 2.38^{b} 17.41^{cd} 11.53c 3.73^{abcd} BS + SM2.73ab 18.57abc 13.95ab 259.21ab 14.28ab BS + VAM3.90ab 19.05ab 271.67ab 2.83a 3.86^{abc} 18.60abc 268.23ab SM + VAM 2.80^{a} 14.48a 3.73abcd 18.44^{abc} 2.73^{ab} 13.78ab 254.63ab BS + PAU consortium 3.71abcd 18.29abcd 13.55ab 247.66abc SM + PAU consortium 2.63^{ab}

3.99a

0.36

19.64a

1.36

Table 3 Effect of different microbial inoculants on fruit characteristics and cumulative yield of strawberry plant

Treatment details are given under Materials and Methods.

 2.85^{a}

0.39

BS + SM + VAM + PAU consortium

LSD (*P*≤0.05%)

+ VAM + PAU consortium significantly enhanced the fresh weight (14.9 g), dry weight (4.3 g), length (21.57 cm) and volume (8.4 cc) of strawberry root. The addition of biostimulants (contains humic acid, amino acids, thiamine etc.) + AMF+ *Bacillus* spp. to the root zone of apple seedlings enhanced the length of roots, fresh and dry weight of roots as compared to non-inoculation (Thomas *et al.* 2022). In banana, root length, root volume and root dry weight was improved by the application of N_2 (33%) + *Azospirillum brasilense* Sp7 (Mia *et al.* 2010).

Fruit parameters and yield: A significant improvement in fruit characteristics was recorded by the inoculation of strawberry plants with various microbial inoculants. The maximum fruit length (3.71 cm), fruit diameter (2.85 cm) and fruit weight (19.64 g) were observed in Burkholderia seminalis + Stenotrophomonas maltophilia + VAM + PAU consortium inoculated strawberry plants. The strawberry plants treated with Stenotrophomonas maltophilia + VAM inoculation resulted in a maximal number of fruits/plant (14.48) (Table 3). Cheng et al. (2022) studied the navel orange plants that were inoculated with Diversispora versiformis (AMF) or Diversispora spurca (AMF) or Piriformospora indica showed increased fruit weight and number of fruits over the non-inoculation. Kumar et al. (2019) concluded, the total number of strawberry fruits (20.44) and fruit weight (17.5 g) were maximum in the application RDF (75%) + Azospirillum (2 g/plant) + soil application of 25% potassium + PSB (2 g/plant). According to Dey et al. (2005), Phosphobactrin and VAM were effective in improving the diameter, length, and weight of guava fruit because they have the capability to improve availability of plant nutrients like phosphorus etc. by solubilizing them.

The cumulative marketable fruit yield of the strawberry/plant was found maximal 277.86 g in *Burkholderia* seminalis + Stenotrophomonas maltophilia + VAM + PAU consortium treated plants of Winter Dawn over the control

(194.10 g). Similar result was revealed by Pandit *et al.* (2013), the application of VAM (12 kg/ha) in strawberry showed enhanced vegetative growth (34.37%), and fruit yield 41.63% higher than the control. The fruit yield of strawberry/plant was enhanced by 25% over the control with the application of 75% nitrogen + *Methylobacterium symbioticum*, it might be due to the enhancement in production of PGR and nitrogen fixation (Vera *et al.* 2023).

14.18^{ab}

1.68

277.86a

33.12

In conclusion the strawberry plants inoculated with different microbial inoculants showed improvement in plant growth characteristics, root characteristics and cumulative marketable yield over the control whereas in terms of combination of different microbial inoculants, the plants treated with *B. seminalis* + *S. maltophilia* + VAM + PAU consortium was performed better among all the different combinations.

SUMMARY

In the rhizosphere, plants interact with a wide range of microorganisms that play crucial role in several aspects of plant development and defence against biotic and abiotic stresses. The present study was carried out during 2022–23 at Regional Research Station (Punjab Agricultural University, Ludhiana), Bathinda, Punjab to elucidate the role of various microbial inoculants on vegetative growth, yield, and quality of strawberry (Fragaria × ananassa). The results depicted that, strawberry plants treated with Burkholderia seminalis + Stenotrophomonas maltophilia + VAM + PAU consortium (T₁₀) was found best over the control in terms of morphological characteristics, viz. plant height, spread, leaf area, number of leaves, crown diameter, and root characteristics of plant. Fruit characteristics like fruit weight, fruit length, and fruit diameter were improved by inoculating the plants with Burkholderia seminalis + *Stenotrophomonas maltophilia* + VAM + PAU consortium. The cumulative marketable yield of both strawberry cultivar winter dawn was enhanced by inoculating the plant with different microbial inoculants but found maximum in *Burkholderia seminalis* + *Stenotrophomonas maltophilia* + VAM + PAU consortium.

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