

## Efficacy of the CSR bio-inoculant and organic amendments in cultivating strawberry cv. Chandler in salt-affected soils

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### Abstract

Strawberry (*Fragaria × ananassa* Duch.), occupies a significant place in fruit industry, since it is cultivated both in plains as well as in hills. The fruit stands out for its organoleptic and nutraceutical properties and is appreciated around the world for its economic returns. Despite their high sensitivity to marginal and salt affected soil, it has a potential, as fruit crops perform well in salt-affected soils when grown with the aid of innovative interventions. The current study is one such attempt where a CSR bio-inoculant comprising of salt tolerant and growth promoting rhizobacterial bio-consortia on a unique media developed at ICAR-Central Soil Salinity Research Institute, Regional Station, Lucknow was assessed for its field efficacy with enrichment of different organic amendments. The field experiment was conducted during 2018-19 at the Horticulture Research Farm of the Dept. of Horticulture at Babasaheb Bhimrao Ambedkar University, Lucknow, India, on Strawberry cv. Chandler grown on partially reclaimed alkali soil of central Uttar Pradesh. The study was laid out in RBD with twelve treatment combinations and control replicated thrice. The results show that runners treated with CSR bio-inoculant and applied with FYM, municipal solid waste compost (MSWC), and paddy straw mulching have a significant influence on fruit yield and physicochemical properties. The maximum fruit length, fruit diameter, fruit size, fruit weight, fruit dry weight, fruit volume, fruit yield/plant (131.06g), and fruit yield/ha (194.16q) were all statistically significant over the control in the treatment with 75% NPK+ MSWC + Mulch (Paddy straw) + CSR bio-inoculant. The experimental findings revealed that treatment with CSR bio-inoculant and 75% NPK+MSWC+paddy straw mulch could increase the yield of strawberry cv. Chandler under salt stress conditions without hampering the soil fertility status, particularly in the subtropical region of the country.

**Key words:** Strawberry, organic, FYM, municipal solid waste compost, CSR bio-inoculant.

### Introduction

The strawberry is not really a berry, but an achene or “false” fruit. The cultivated strawberry (*Fragaria × ananassa* Duch.) belongs to the family Rosaceae. In the 18th century (1714 & 1759), it arose in Europe as a chance cross between two American native diploid species viz., *Fragaria chilioensis* Duch. and *Fragaria virginiana* Duch. (Singh *et al.*, 2015). It is an herbaceous crop with a healthy prostrate growth habit, which behaves as an annual in the sub-tropical region and perennial in the temperate region (Beer *et al.*, 2017). It is cultivated in about 75 countries in plain and in hilly areas up to an elevation of 3000 m from MSL in humid or dry cold regions (Singh *et al.*, 2008) with an assured irrigation facility. Strawberry is used as a fresh fruit rich in vitamin C and ellagic acid, which has anti-cancerous properties. It is a valuable food in the diet of millions of people around the globe. It is worthy to note that strawberry gives high returns in a short time among all the fruits.

Fruits are attractive with distinct pleasant aroma and flavor and a sweet taste, deliciousness, softness, and rich source of vitamins, minerals and nutrients. (Sharma and Sharma, 2003), consumed as dessert and have a special demand by the fruit processing industries to prepare canned strawberries, jam, jelly, ice cream, and beauty products (Hughes *et al.*, 1969). The taste of fruit mainly depends on three different compounds: sugars, acids, and aromatic compounds. The strawberry fruit contains 0.55%

total sugar and 0.90% to 1.85% acidity, prominent as malic and citric acid. As reported by Singh *et al.* (2017), 100g fresh fruit contains, edible portion (96%), moisture (87.8%), protein (0.7%), fat (0.2%), fiber (1.1%), carbohydrate (9.8%), minerals (0.4%), vitamin A (30 iu/100 g), thiamine (0.03 mg/100 g), riboflavin (0.07 mg/100 g), nicotinic acid (0.2 mg/100 g), ascorbic acid (55/100 g) and calories (44/100 g).

Salt affected soils contributes for the major category under the waste lands occupying an extensive area in the world and in India (6.73 mha) as well, presenting serious impediments to crop production (Gupta and Abrol, 2000). High alkalinity and high exchangeable sodium percentage (ESP>15) of the soil with minimal biological activity hinders the growth of commercial fruit crops even after reclamation using gypsum technology (Damodaran *et al.*, 2013). Possible strategy to counteract the adverse effect of salinity is to exploit the avenues of bio-agents or bio-inoculants that are harnessed from the salt stress rhizosphere (Egamberdieva, 2008). CSR bio-inoculant is a biosmart microbial consortia comprising of the salt tolerant and growth promoting bacterial isolates CSR-M16 (*Bacillus licheniformis*), CSR-A11 (*Lysnibacillus fusiformis*) and CSR-A16 (*Lysnibacillus sphaericus*) identified with higher vigor index and growth promotion in sodic soil conditions upto soil pH 9.30 cultured on CSR patent protected liquid media (Damodaran *et al.*, 2019).

The current study aimed to evaluate CSR bio-inoculant consisting

of salt-tolerant rhizobacterial consortia for field efficacy with enrichment of various organic amendments.

## Materials and methods

The present study was carried out at the Horticulture Research Farm of the Dept. of Horticulture Babasaheb Bhimrao Ambedkar University, Lucknow, Uttar Pradesh during the winter season of 2018-2019. The soil pH of the experimental field ranged from 8.9 to 9.0. Twelve treatment combinations with three replications were tested in Randomized Block Design. The treatments involving CSR bio-inoculant was made by addition of organic amendments enriched with the bio-inoculant @ 1 percent drenching, 24 hrs before application of the organic amendment to the plots. The observations were recorded on five fruits having uniform shape and size, randomly harvested from each treatment. Five morphological or physical characters of fruits and yield (Table 1) were used to characterize and describe the 12 treatments of strawberry. Twelve physico-chemical attributes constitutes were studies of fruit and investigation was analyzed statistically.

**pH determination of fruit juice:** The pH of the each fruit sample was measured with a digital pH meter. The pH meter was standardized against standard buffer solution before use. Fruit juice of each sample was taken in a clean beaker and its pH was measured at room temperature.

**Total soluble solids (°Brix):** Pulp was taken from five randomly selected fruits from each treatment and prepared a composite sample and crushed them for juice extraction. TSS was measured with the help of a digital hand refractometer (0-32 range). A drop of fruit juice was put on lance of digital hand refractometer and the value was noted. Values thus, obtained were expressed in degree brix (°Brix).

**TSS/Acid ratio:** TSS/acid ratio was calculated by dividing total soluble solids (TSS °Brix) by acidity %.

**Titrateable acidity (%):** Titrateable acidity was determined by titration in terms of citric acid with NaOH 0.1 N where one hundred gram fruit pulp was crushed in a blender and juice was filtered with muslin cloth. 10 ml. of fruit juice sample was taken and make up volume up to 100 mL by adding distilled water. Taken 10 mL of this aliquot and titrated against 0.1 N NaOH using few drops of 1% phenolphthalein indicator till the pink color was appeared (end-point persists).

**Ascorbic acid (mg/100 g):** Ascorbic Acid was determined by using 2, 6- dichlorophenol-indophenol through visual titration method. 10 mL fruit juice sample was taken and volume was made up to 100 mL by adding distilled water with 3% metaphosphoric acid (HPO<sub>3</sub>) and filtered with filter paper. 10 mL aliquot was titrated against standard dye (2, 4- Dichlorophenol indophenols) to obtain a pink colour was observed which persisted for at least 15 seconds.

**Sugars:** Total, reducing and non reducing sugars were estimated in 10 g fresh fruit pulp as suggested by Rangana (1997).

**Anthocyanin (mg/100g):** The Anthocyanin was determined by the method as suggested by Rangana (1997). Dilute 10 mL of juice to 50 mL. with 0.1N HCL and allow equilibrating in the dark for one hour. Record the absorbance (O.D.) at 510 nm.

Total O.D./100 mL = O.D.X Volume made up X 100 / mL of juice taken  
Total anthocyanin (mg /100 ml) = Total O.D./100ml / 87.3.

## Results and discussion

The data regarding the different physico-chemical attributes and yield expressed in (Table 1 and 2) were observed at clearly indicate that the application of integrated sources of nutrients and organics *viz.*, farm yard manure (FYM), municipal solid waste compost (MSWC) and bio-inoculant significantly affect the physico-chemical attributes.

**Physical characters of fruits:** The physical characters *viz.*, fruit length, fruit diameter, fruit size, fresh fruit weight, dry weight, fruit volume and fruit specific gravity are constituents for assessing physical characters of the fruits of different treatments of strawberry and mean data were recorded and expressed in (Table 1 & 2). Fruit length ranged from 2.18 to 3.83 cm and maximum (3.83 cm) was recorded in treatment T<sub>9</sub>, followed by T<sub>1</sub>:100% NPK + Mulch (Paddy straw) (3.70 cm). The minimum (2.18cm) fruit length was recorded in T<sub>0</sub> (control). Fruit diameter ranged from 1.46cm to 2.81cm. Maximum fruit diameter was observed in treatment with T<sub>9</sub> (2.81 cm), followed by T<sub>8</sub> (2.74cm). The minimum fruit diameter was recorded in untreated *i.e.* T<sub>0</sub> (1.46 cm). Fruit size ranged from 3.19 to 9.67 cm<sup>2</sup>. Maximum fruit size (9.67 cm<sup>2</sup>) was found in treatment with T<sub>9</sub>, followed by T<sub>1</sub>:100% NPK + Mulch (Paddy straw) (9.38 cm<sup>2</sup>). The minimum fruit size was recorded in control (3.19cm<sup>2</sup>). Fruit weight ranged from 5.94 to 13.10g. Maximum fruit weight (13.10g) was found in treatment with T<sub>9</sub>, followed by T<sub>8</sub> (12.74g). The minimum

Table 1. Assessment of FYM, MSWC and mulching with bio-inoculant (PSB) on yield and physical attributes of fruits in strawberry

Treatments	Length of fruit (cm)	Diameter of fruit (cm)	Size of fruit (cm <sup>2</sup> )	Fruit yield / plant (g)	Fruit yield / ha. (q)
T <sub>0</sub> [Control]	2.18	1.46	3.19	35.14	52.06
T <sub>1</sub> [100% NPK + Mulch (Paddy straw)]	3.70	2.54	9.38	86.96	128.83
T <sub>2</sub> [75% NPK + Mulch (Paddy straw)]	3.39	1.92	7.00	81.22	120.32
T <sub>3</sub> [50% NPK + Mulch (Paddy straw)]	3.04	1.84	5.80	59.10	87.55
T <sub>4</sub> [75% NPK + FYM + Mulch (Paddy straw)]	3.22	2.24	7.22	95.50	141.49
T <sub>5</sub> [75% NPK + MSWC + Mulch (Paddy straw)]	3.23	2.35	7.59	103.64	153.55
T <sub>6</sub> [50% NPK + FYM + Mulch (Paddy straw)]	2.94	2.22	6.52	82.93	122.86
T <sub>7</sub> [50% NPK + MSWC + Mulch (Paddy straw)]	2.94	1.88	5.52	87.14	129.09
T <sub>8</sub> [75% NPK + FYM + Mulch (Paddy straw) + Bio-inoculant]	3.59	2.74	8.83	125.54	185.99
T <sub>9</sub> [75% NPK+MSWC + Mulch (Paddy straw)+ Bio-inoculant]	3.83	2.81	9.67	131.06	194.16
T <sub>10</sub> [50% NPK+ FYM + Mulch (Paddy straw) + Bio-inoculant]	3.33	2.25	7.49	116.37	172.40
T <sub>11</sub> [50% NPK+MSWC+Mulch (Paddy straw) + Bio-inoculant]	3.44	2.16	7.43	125.10	185.33
CD at 5%	0.701	N/A	1.865	20.677	30.632
SEm±	0.238	0.215	0.632	7.005	10.377

Table 2. Assessment of FYM, MSWC and mulching with bio-inoculant (PSB) on physico-chemical attributes in strawberry cv. Chandler (2018-19)

Treatments	Fresh fruit weight (g)	Volume of fruit (mL)	Specific gravity	Dry fruit weight (g)	pH of fruit juice	TSS (°Brix)
T <sub>0</sub> [Control]	5.94	6.28	0.946	0.57	3.86	6.29
T <sub>1</sub> [100% NPK + Mulch (Paddy straw)]	9.43	10.00	0.943	0.90	4.26	9.85
T <sub>2</sub> [75% NPK + Mulch (Paddy straw)]	9.40	9.05	1.039	0.90	4.13	9.79
T <sub>3</sub> [50% NPK + Mulch (Paddy straw)]	8.26	8.02	1.030	0.79	4.15	9.57
T <sub>4</sub> [75% NPK + FYM + Mulch (Paddy straw)]	9.99	9.70	1.031	0.95	4.73	10.22
T <sub>5</sub> [75% NPK + MSWC + Mulch (Paddy straw)]	10.50	10.25	1.024	1.00	4.46	10.54
T <sub>6</sub> [50% NPK + FYM + Mulch (Paddy straw)]	9.32	9.21	1.011	0.89	4.19	9.38
T <sub>7</sub> [50% NPK + MSWC + Mulch (Paddy straw)]	9.00	8.65	1.041	0.86	4.27	9.37
T <sub>8</sub> [75% NPK + FYM + Mulch (Paddy straw) + Bio-inoculant]	12.74	12.26	1.039	1.21	4.89	12.28
T <sub>9</sub> [75% NPK+MSWC + Mulch (Paddy straw)+ Bio-inoculant]	13.10	12.64	1.036	1.25	4.86	12.05
T <sub>10</sub> [50% NPK+ FYM + Mulch (Paddy straw) + Bio-inoculant]	12.03	11.60	1.036	1.15	4.90	11.53
T <sub>11</sub> [50% NPK+MSWC+Mulch (Paddy straw) + Bio-inoculant]	12.58	12.19	1.032	1.20	4.73	10.47
CD at 5%	1.947	1.865	0.026	0.187	0.407	1.015
SEm±	0.660	0.632	0.009	0.063	0.138	0.344

fruit weight was recorded in the control (5.94g). Fruit dry weight ranged from 0.57 to 1.25g. Maximum fruit weight (1.25g) was found in treatment with T<sub>9</sub>, followed by T<sub>8</sub> (1.21g). The minimum fruit weight was recorded in control (0.57g). Fruit volume ranged from 6.28 to 12.64cc. Maximum fruit volume was observed in treatment with T<sub>9</sub> (12.65cc), followed by T<sub>8</sub> (12.26cc). The minimum fruit volume was recorded in the control (6.28 cc). Fruit specific gravity ranged from 0.943 to 1.041. Maximum fruit specific gravity was recorded in the treatment with T<sub>7</sub>; 50% NPK + MSWC + Mulch (Paddy straw) (1.041), followed by T<sub>8</sub> (1.039). The minimum fruit specific gravity was recorded in T<sub>1</sub> (0.943). Berry size and weight are highly related with the dry matter content and balance level of hormone. Bio-inoculants like Azospirillum and Pseudomonas tend to accumulate the dry matter and aid in their translocation (Kachot *et al.*, 2001), it also favour synthesis of different growth regulators (Awasthi *et al.*, 1998),

**Fruit yield:** Significantly high fruit yield/plant (131.06g) was observed in treatment T<sub>9</sub> followed by T<sub>8</sub> (125.54g). The minimum fruit yield/plant was recorded under control (35.14g). Treatment T<sub>9</sub> had maximum fruit yield/ha (194.16q), followed by T<sub>8</sub> (185.99q). The minimum fruit yield/ha was recorded under control (52.06q). Acetobacter application along with vermicompost tend to increase the yield of strawberry variety Chandler (Mishra and Tripathi, 2012). Similar findings were reported by Kadlag *et al.* (2007) in tomato and Nowsheen *et al.* (2006) in cv. Senga

Sengana of strawberry. The increase in yield may be due to the increase in the uptake of phosphorous and potassium from the soil due to the application of the CSR bio-inoculant comprising of phosphorous solubilizers and auxin inducers. *Lysnibacillus fusiformis* and *Lysnibacillus sphaericus* were found to possess the plant growth promoting properties of phosphorous solubilizers and IAA production (Damodaran *et al.*, 2019). The bacterial isolate CSR-M16 isolated from the rhizosphere of the mango rootstock ML-2 increase the nutrient uptake of the tomato plants cultivated in the farming system land modification models under the waterlogged sodic soils (Damodaran *et al.*, 2021). CSR bioformulation application can effectively reduce the need for additional 20% nitrogen required under DSR (Direct Seeded Rice) conditions in rice (Parveen kumar *et al.*, 2021).

**Biochemical attributes of the fruits:** The fruit quality parameters of strawberry were significantly influenced by the application of bio-inoculants with the organic amendments and mulching. The pH, total soluble solids, acidity, TSS/Acid ratio, ascorbic acid, reducing sugar, non-reducing sugar and anthocyanin constitute the important chemical constituents for assessing the fruit quality of the fruits of different treatments of strawberry and mean data were recorded and expressed in (Table 2 & 3). The pH of the fruit juice of strawberry ranged from 3.86 to 4.90. The maximum (4.90) pH range of strawberry fruit was recorded in T10 followed by T8 (4.89). The minimum amount of pH was recorded in the control (3.86). The total soluble solids ranged from 6.29 to

Table 3. Assessment of FYM, MSWC and mulching with bio-inoculant (PSB) on biochemical attributes in strawberry cv. Chandler (2018-19)

Treatments	Titrateable acidity (%)	TSS/Acid ratio	Ascorbic acid / (mg/100g)	Reducing sugar (%)	Non-reducing sugar (%)	Anthocyanin (mg/ 100 g)
T <sub>0</sub> [Control]	0.82	7.67	69.51	2.62	2.13	30.79
T <sub>1</sub> [100% NPK + Mulch (Paddy straw)]	0.78	12.68	68.16	2.87	2.25	34.92
T <sub>2</sub> [75% NPK + Mulch (Paddy straw)]	0.76	12.84	64.80	3.15	2.14	35.16
T <sub>3</sub> [50% NPK + Mulch (Paddy straw)]	0.79	12.07	65.09	3.24	2.17	31.22
T <sub>4</sub> [75% NPK + FYM + Mulch (Paddy straw)]	0.67	15.25	57.93	4.03	3.16	41.43
T <sub>5</sub> [75% NPK + MSWC + Mulch (Paddy straw)]	0.71	14.78	59.85	4.60	2.25	46.06
T <sub>6</sub> [50% NPK + FYM + Mulch (Paddy straw)]	0.78	12.08	65.49	3.03	2.13	36.15
T <sub>7</sub> [50% NPK + MSWC + Mulch (Paddy straw)]	0.79	11.92	63.26	2.82	2.38	36.55
T <sub>8</sub> [75% NPK + FYM + Mulch (Paddy straw) + Bio-inoculant]	0.63	19.39	54.41	4.70	3.56	43.23
T <sub>9</sub> [75% NPK+MSWC + Mulch (Paddy straw)+ Bio-inoculant]	0.65	18.45	55.96	4.18	3.26	43.70
T <sub>10</sub> [50% NPK+ FYM + Mulch (Paddy straw) + Bio-inoculant]	0.66	17.41	55.06	4.04	3.16	41.08
T <sub>11</sub> [50% NPK+MSWC+Mulch (Paddy straw) + Bio-inoculant]	0.74	14.09	58.65	3.90	3.15	38.75
CD at 5%	0.025	1.475	3.418	0.425	0.184	2.956
SEm±	0.008	0.500	1.158	0.144	0.062	1.001

12.28°Brix. The maximum (12.28 °Brix) total soluble solids was recorded in T<sub>8</sub>, followed by T<sub>9</sub> (12.05 °Brix) (PSB), while minimum total soluble solids was recorded in control (6.29 °Brix). The acidity ranged from 0.63 to 0.82%. The minimum (0.63%) acidity was noted in T<sub>8</sub> treated plant. While maximum (0.82%) acidity was recorded under control. TSS/Acid ratio ranged from 7.67 to 19.39%. The maximum (19.39%) TSS/A Hughes, H.M., J.B. Duggan and M.G. Banwell, 1969. Strawberry Bull. 95 HMSO, 10, 6d Min. Agric. Fisheries and Food, U.K. cid ratio was recorded in T<sub>8</sub>, followed by T<sub>9</sub> (18.45%). The minimum TSS/Acid ratio was recorded in control (7.67%). Ascorbic acid, the major constituent of strawberry ranged from 54.41 to 69.51 mg/100g of fruit pulp. Increase in the TSS and total sugars of strawberry fruits due to inoculation of free-living nitrogen-fixing bacteria might be due to enhanced supply of nutrients by the bio-inoculants during the crop growth period. This increased the leaf area with higher synthesis of assimilates due to enhanced rate of photosynthesis facilitating the mobility of photosynthetic products from leaves to developing fruits, thereby increasing TSS and total sugars (Singh and Singh, 2006)

The maximum (69.51mg/100g) amount of ascorbic acid was recorded in control followed by T<sub>1</sub> (68.16 mg/100 g). The minimum amount of ascorbic acid was observed in T<sub>8</sub> (54.41mg/100g). Reducing sugar ranged from 2.62 to 4.70%. The maximum (4.70%) reducing sugar was recorded in T<sub>8</sub>, followed by T<sub>5</sub> (4.60%). The minimum reducing sugar (2.62%) was recorded in control. Non-reducing sugar ranged from 2.13 to 3.56%. The maximum (3.56%) non-reducing sugar was recorded in T<sub>8</sub>, followed by T<sub>9</sub> (3.26%). The minimum reducing sugar (2.13 %) was recorded in control. Anthocyanin, the strawberry's major component, ranged from 30.79 to 46.06mg/100g of fruit pulp. The maximum (46.06 mg/100 g) amount of anthocyanin was recorded in T<sub>5</sub> followed by T<sub>9</sub> (43.70mg/100g). The minimum amount (30.79 mg/100g) of anthocyanin was recorded in T<sub>0</sub>. Increased translocation of assimilates from leaves to the developing fruits are highly correlated with endogenous levels of hormones. Bio-inoculants like *Azospirillum* and phosphate solubilizing bacteria of *Bacillus sp.* are known to favour the synthesis of different growth regulators like IAA, GA (Awasthi *et al.*, 1998). Similar results with significant increase in the fruit quality of the strawberry was earlier reported by Hazarika *et al.* (2015).

Our studies indicated that application of the CSR bio-inoculant comprising of consortia of plant growth promoting bacteria isolated from the salt affected rhizosphere tend to increase the yield and quality parameters of the strawberry var. chandler in the salt affected soils when used along with the organic amendments like municipal solid waste. The use of the CSR-bioinoculants with soil amendments shall enable the cultivation of the strawberry in salt affected soils.

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