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Reproductive Growth, Yield, and Quality of Strawberry Cv. Camarosa Intensify by Integrated Bio-Stimulants Application

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Abstract

Purpose: The study was done in the naturally ventilated polyhouse at Lovely Professional University, Phagwara, Jalandhar, Punjab, in the department of horticulture. The goal was to find out how bio-fertilizer and organic manure affected the growth, yield, and quality of Camarosa strawberries that were grown organically. **Research Method:** The experiment was laid out in a completely randomized design with nine treatments and three replications. Three manure (vermicompost, neem cake, cocopeat) and 2 biofertilizers (Azobactor and PSB) were applied individually and in composite form of both biofertilizers mixed with individually in each manure and all manure, to compare their individual effect and synergic effect on strawberry plant growth and fruit quality. **Findings:** The results revealed that the highest fruit productivity was obtained from a combination of Vermicompost @50g plant⁻¹, Neem cake @50g plant⁻¹, Cocopeat @50g plant⁻¹, Azotobacter @1g plant⁻¹, and PSB @1g plant⁻¹ (VC+NC+CP+AZ+PSB), while the largest fruits were obtained from Neem Cake @100g plant⁻¹. No significant differences were found between the different treatments for titratable acidity. The highest T.S.S., ascorbic acid, and total sugar of strawberry fruit were recorded from VC+NC+CP+AZ+PSB treated plants. **Value:** The integrated application of bio-stimulants increases the yield and quality of fruits over the individual treatment. The study is useful for the farmers to increase the yield of fruits with natural application.

Keywords: Bio-fertilizer, Organic manure, Vermicompost, Strawberry, Productivity.

Introduction

Strawberry, scientifically known as *Fragaria × ananassa* Duch., stands as a beloved soft berry fruit enjoyed globally. This fruit emerges from a hybrid lineage, a result of crossbreeding two American species: *Fragaria chiloensis* Duch. and *Fragaria virginiana* Duch. (Singh et al., 2015; Urso et al., 2015). Initially traced back to 18th century Europe, strawberries spurred the development of diverse varieties across various countries throughout the 19th century. Most cultivated strawberry types exhibit octaploid characteristics, featuring a chromosome count of $2n = 8x = 56$. Within the Rosaceae family's *Fragaria* genus, consisting of 20 wild species,

three are natural hybrids. The well-recognized modern cultivated strawberry, *Fragaria × ananassa* Duch., has its origins spanning Asia, North America, Europe, and South America (Li et al., 2011).

The strawberry, an herbaceous fruit crop, behaves as an annual in subtropical regions and a perennial in cooler climates. Its popularity arises from its pleasing taste, appealing flavor, vivid color, convenience, high yield, and quality (Khalid et al., 2020). Of note are the dominant constituents in its phytochemical makeup: anthocyanins, phenolic compounds, and ascorbic acid. These components confer various medicinal benefits, including antioxidative, anticarcinogenic, anti-inflammatory, and anti-neurodegenerative properties (Fernandes et al., 2012; Urso et al., 2015; Domingues et al., 2018).

The utilization of organic fertilizers and biofertilizers holds a promising avenue for enhancing agricultural inputs. An instance of such is vermicompost, a result of the symbiotic interaction between earthworms and microorganisms, leading to the biodegradation and stabilization of organic materials. This process yields a nutrient-rich product containing vital elements such as nitrates, exchangeable calcium, phosphates, and soluble potassium that are readily accessible to plants (Kumar et al., 2018; Negi et al., 2021). In a similar vein, biofertilizers comprise living microorganisms naturally present in the soil or plant rhizosphere. These microorganisms inherently foster plant growth (Negi et al., 2011; Sawana et al., 2014; Borriss, 2017), offering an array of positive effects on both soil health and the host plant. This natural approach presents distinct ecological advantages over inorganic chemicals, contributing to increased soil fertility and biological activity, ultimately boosting crop production (Shen et al., 2011). This study aimed to evaluate the results obtained from an experiment that explored the effects of different treatments utilizing biofertilizers and organic fertilizers. The focus was on understanding how these treatments impact the reproductive growth, quality, and overall productivity of strawberry fruits.

Materials and Methods

Experimental Site

The research was conducted within a naturally ventilated polyhouse located at Lovely Professional University's Department of Horticulture in Phagwara, Jalandhar, Punjab, during the period of 2021–2022. Camarosa strawberry fruit runners for the experiment were sourced from AKS Strawberry in Solan, Himachal Pradesh. Organic manures such as vermicompost, neem cake, and cocopeat were procured from a local fertilizer store in Jalandhar. The university's store provided the fertilizers (Azotobacter and PSB) necessary for the treatments. The planting of strawberry runners occurred in November 2021 on a bed measuring 40 x 2 m², accompanied by an underlying drip irrigation system. The bed was divided into ten equal sections, each spanning 4 meters. Plant spacing was set at 30 cm between rows and 45cm between individual plants. The temperature and

humidity of the polyhouse during the experiment were observed at 20 ± 3 °C and 82 ± 4 %, respectively. The soil was sandy loamy with an alkaline pH of 7.65, 1.00 dS m⁻¹ electrical conductivity, 0.40% organic carbon, 0.029 g/kg nitrogen, 0.002 kg/ha phosphorus, and 0.024 kg/ha potassium. The fruit quality data were recorded after harvesting, while yield related parameters were recorded at a weekly interval, and the sum was calculated at the end of April.

Treatment Details

The experiment encompassed the application of organic manures and biofertilizers to the soil. The experiment was designed with eight treatments and one control. Organic manures, specifically vermicompost, neem cake, and cocopeat, were employed, along with the utilization of biofertilizers like PSB and Azotobacter. Employing a completely randomized design, the experiment comprised nine distinct treatments and a control group (Table 1), with each treatment replicated three times.

Table 01: Details of the soil application treatments for Camarosa strawberry

Notations	Treatments	Composition(g plant ⁻¹)
T0	Control	0
T1	Vermicompost (VC)	100
T2	Neem Cake (NC)	100
T3	Cocopeat (CP)	100
T4	<i>Azotobacter chroococcum</i> (AZ)	2.5
T5	Phosphate Solubilizing Bacteria (PSB)	2.5
T6	VC+AZ+PSB	100+1+1
T7	NC+AZ+PSB	100+1+1
T8	CP+AZ+PSB	100+1+1
T9	VC+NC+CP+AZ+PSB	50+50+50+1+1

Statistical Detail

The experiment was conducted under a one factor ANOVA randomized block design with three replications. The data was observed, and the mean value was calculated with a standard error by the Duncan test using the statistical tool SPSS.

Results and Discussion

The productivity was recorded to be maximum (8.62 q/ha) in VC+NC+CP+AZ+PSB (Vermicompost @50g plant⁻¹ + Neem cake @50gplant⁻¹ + Cocopeat @50gplant⁻¹ + *Azotobacter* @1gplant⁻¹ + PSB @1gplant⁻¹) treated plants (Figs. 1 and 1a). Whereas, highest fruit set (96.68 %) was recorded in plants of NC+AZ+PSB (Neem cake @100g plant⁻¹ + *Azotobacter* @1g plant⁻¹ + PSB @1g plant⁻¹). However, the combination showed highest productivity but the NC individually have better result followed by PSB and VC. Whereas, a small variation

was observed among the treatments for fruit set as compared to control (Figure 1). Organic matter, phosphorous availability, and total organic nitrogen percentage play an important role in strawberry through the formation of carbohydrates, and composted manure accelerates biological activity in soil, helps fortify soil fertility and enhances yield (Sahana *et al.*, 2020).

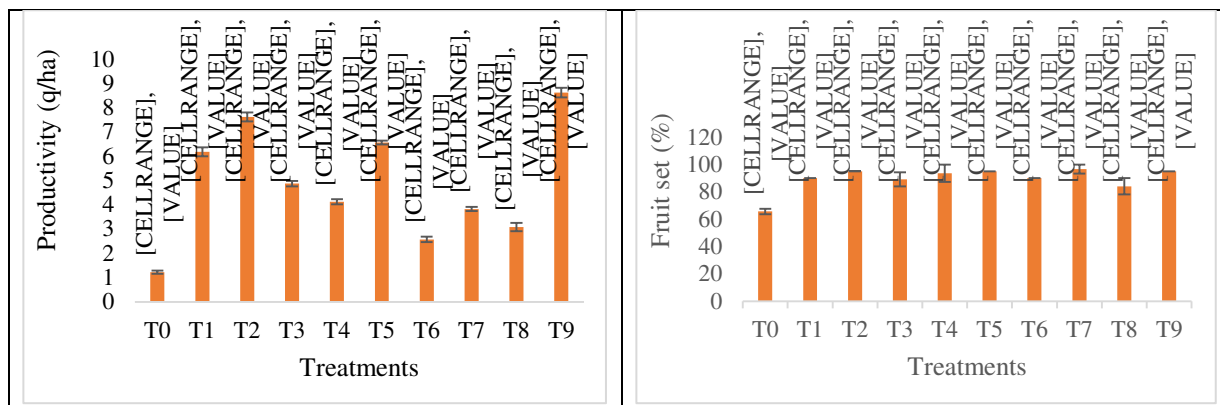


Figure01:Trend of changes in productivity and fruit set of Camarosa strawberry due to different organic manure and biofertilizers. Letters indicate the significance level at 5% probability.

The synergistic effects on improving soil fertility, nutrient availability, and plant health create a conducive environment for robust reproductive growth in strawberry plants. Neem cake, azotobacter, and PSB enhance nitrogen and phosphorus availability in plants, a critical nutrient for flowering and fruit development, thus leading to early fruit setting in plants (Srivastav *et al.*, 2018).





Figure 1a Strawberry growth after the application of organic manure and

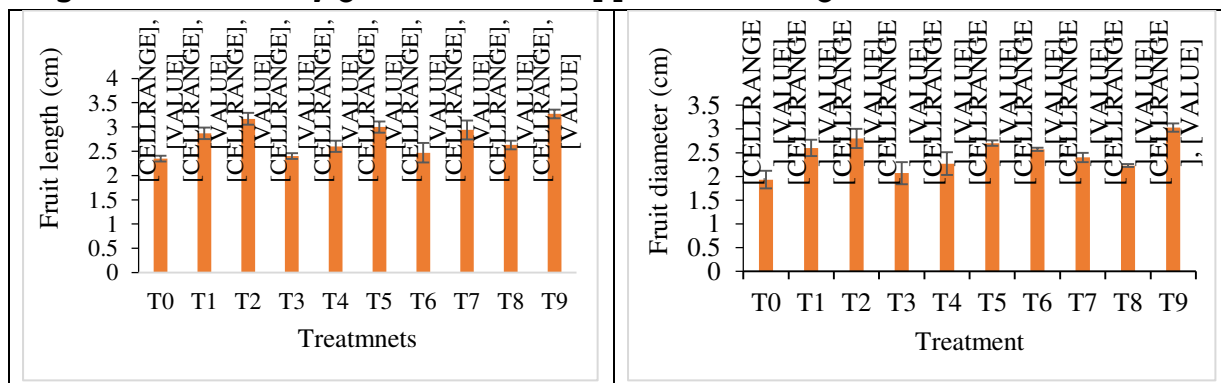


Figure 02: Changes in fruit length and diameter of Camarosa strawberry due to different organic manure and biofertilizers. Letters indicate the significance level at 5% probability.

The maximum fruit length (3.27 cm) and fruit diameter (3.03 cm) of strawberry fruits were observed under treatment T9 (Vermicompost @50 g plant⁻¹ + Neem cake @50 g plant⁻¹ + Cocopeat @50 g plant⁻¹ + Azotobacter @1 g plant⁻¹ + PSB @1 g plant⁻¹) in Figure 2. The increase in fruit length and fruit diameter contributed to better fillings of fruits; this might be due to the uptake of more balanced nutrients, which led to superior metabolic activities in the plant that

ultimately led to high carbohydrate and protein synthesis. Khalid et al. (2013) and Martinsson et al. (2009) have both reported similar observations in strawberries. Biofertilizers and organic fertilizers worked together to speed up the loading and mobilization of photo assimilates into fruits. This led to faster cell division and expansion, which made the fruits bigger and longer. Furthermore, organic fertilizers improved the plant's ability to make food, make growth regulators, and move hormones around, all of which helped the fruit grow (Yadav et al., 2010; Khalid et al., 2013).

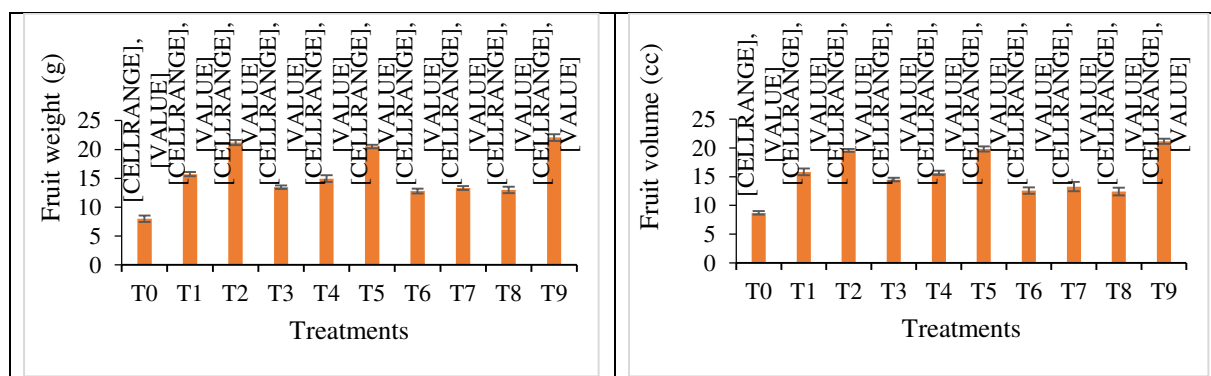


Figure03:Trend of changes in fruit weight and volume of Camarosa strawberry due to different organic manure and biofertilizers. Letters indicate the significance level at 5% probability.

Among various treatments, the maximum fruit weight (22.07 g) and fruit volume (21.17 cc) were recorded in the treatment of combined soil application of biofertilizers and organic fertilizers, i.e., T9 (Vermicompost @50 g plant⁻¹ + Neem cake @50 gplant⁻¹ + Cocopeat @50 gplant⁻¹ + *Azotobacter* @1 gplant⁻¹ + PSB @1 gplant⁻¹), as shown in Figure 3. The concurrent application of biofertilizers (*Azotobacter* and PSB) and organic fertilizers (Vermicompost, Neem cake, and Cocopeat) to the soil resulted in an augmented photosynthetic capacity of the plants, likely contributing to an increased accumulation of dry matter. This increase in dry matter content had a positive effect on the weight and volume of the fruit. This is related to hormone balance, the presence of nitrogen-fixing microorganisms (which are known for storing dry matter), and the efficient movement of nutrients. The symbiotic utilization of biofertilizers and the application of N, P, and K by the plants led to the production of more substantial photosynthates, reflected in higher biomass, which in turn contributed to greater fruit volume due to efficient assimilate translocation to the developing fruits. These findings are in line with earlier research on strawberry cultivation by Yadav et al. (2010), Khalid et al. (2013), Beer et al. (2017), and Neetu and Sharma (2018).

According to Figure 4, the highest specific gravity (1.09 g/ml) was recorded under Neem cake @100 g plant⁻¹ the treatment. The plant efficiently

employed the applied N, P, and K along with bio-fertilizers and organic fertilizers, which lead to the highest photosynthate production with respect to high biomass concentration, and their assimilate translocation to the rising sink leads to the development of a higher specific gravity. Moreover, specific gravity plays a major role in terms of fruit majority as it is a good maturity judging index for fruit crops (Neetu and Sharma 2018).

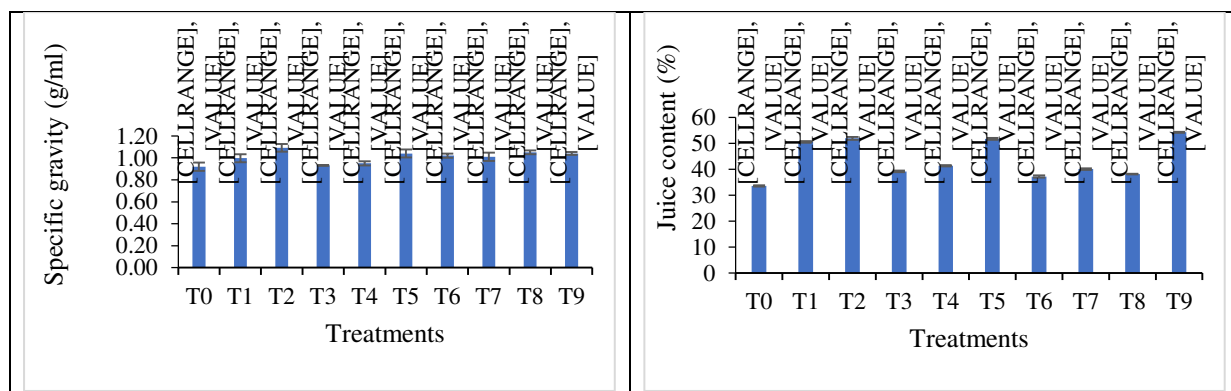


Figure04:Trend of changes in fruit specific gravity and juice content of Camarosa strawberry due to different organic manure and biofertilizers. Letters indicate the significance level at 5% probability.

FromFigure 4, it is evident that the highest juice percentage (54.35%) was recorded under treatment T9 (Vermicompost @50 g plant⁻¹ + Neem cake @50 gplant⁻¹ + Cocopeat @50 gplant⁻¹ + Azotobacter @1 gplant⁻¹ + PSB @1 gplant⁻¹). The collaborative effect of nitrogen and potassium resulting from the combined soil application of organic fertilizers (Vermicompost, Neem cake, and Cocopeat) and biofertilizers (Azotobacter and PSB) resulted in an elevation of juice percentage. This phenomenon is attributed to the efficient utilization of these nutrients, along with others, in the sugar metabolism of strawberry fruits (El-Hamid *et al.*, 2006). These observations align with similar outcomes reported by Kumar *et al.* (2015) in the context of strawberry cultivation.

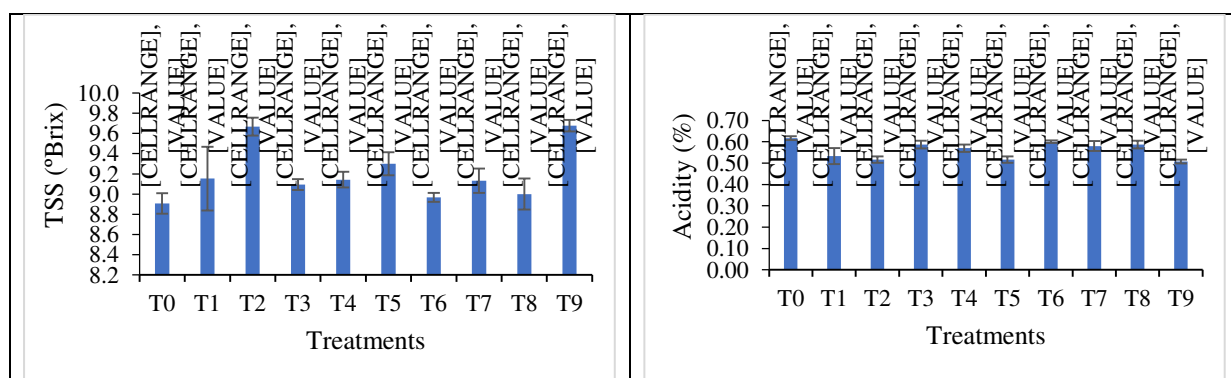


Figure05:Trend of changes in fruit TSS and acidity of Camarosa strawberry due to different organic manure and biofertilizers. Letters indicate the significance level at 5% probability.

Based on the data provided in Figure 5, the maximum TSS was observed under treatment T9 (Vermicompost @50 g plant⁻¹ + Neem cake @50 g plant⁻¹ + Cocopeat @50 g plant⁻¹ + *Azotobacter* @1 g plant⁻¹ + PSB @1 g plant⁻¹). Utilizing a combination of bio-fertilizers, organic fertilizers, and NPK has been found to elevate the total soluble solids (TSS) concentration. This effect is likely attributed to factors such as increased sugar accumulation, ascorbic acid oxidation, and enhanced protein hydrolysis. Notably, the joint application of NPK, *Azospirillum*, and vermicompost further contributes to heightened TSS levels. This rise is because the plant's metabolism quickly changes pectin and starch into soluble parts, sugar moves quickly from the leaves to the developing fruits, and there are more essential micronutrients, macronutrients, carbohydrates, and proteins. These findings are in support of the results recorded by Bhagat and Panigrahi (2020), Singh and Singh *et al.* (2009), Tripathi *et al.* (2015), Ameri *et al.* (2012), and Khalid *et al.* (2013).

According to the investigation results manifested in Figure 5, the titratable acidity value (0.62%) was highest in T0 (control) plants. The reduction in the value of titratable acidity, might be due to a reduction in soil pH, making the soil slightly acidic, thus helping to increase the sweetness of strawberries and the conversion of higher amounts of organic acids and photosynthates into sugars during the fruit ripening stage. These findings are in line with Tripathi *et al.* (2015) and Sharma *et al.* (2019).

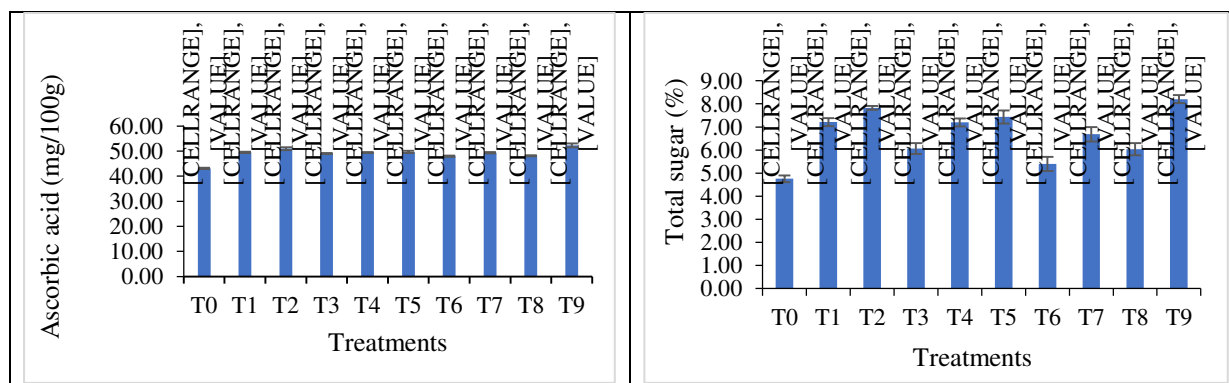


Figure06:Trend of changes in ascorbic acid and total sugar of Camarosa strawberry fruits due to different organic manure and biofertilizers. Letters indicate the significance level at 5% probability.

The data given in Figure 6 make it evident that the ascorbic acid (52.33 mg/100 g) and total sugar (8.20%) content were recorded at their maximum in treatment T9 (Vermicompost @50 g plant⁻¹ + Neem cake @50 g plant⁻¹ + Cocopeat @50 g plant⁻¹ + *Azotobacter* @1 g plant⁻¹ + PSB @1 g plant⁻¹). The high concentration of ascorbic acid might be due to the containing of micro and macronutrients. The fixation of atmospheric nitrogen by microbial inoculants is enhanced. Phosphorus availability is increased, and growth-promoting

substances are secreted, which accelerate biological processes like the synthesis of carbohydrates. These findings agree with the results of Beer *et al.* (2017) and Tripathi *et al.* (2015).

The combined application of biofertilizers (*Azotobacter* and PSB) and organic fertilizers (Vermicompost, Neem cake and Cocopeat) along with NPK, has increased the total sugars content, and this increasement might be due to the quick metabolic conversion of pectin and starch into soluble components and immediate sugar translocation to the developing fruits from leaves. It might also be due to degradation of polysaccharides into monosaccharide's; and increased micronutrients, macronutrients, carbohydrates and protein. Esitkenet *al.*,(2010) stated that as an endogenous factor affects fruit quality, nitrogen absorption may have played a major role in influencing fruit quality, as the roots and stem of the fruit are heavily reliant on carbohydrate during ripening, resulting in high sugar levels in fruit. These findings are in conformity with the findings of Lata *et al.*,(2018), Ahmad and Mohammad (2012), Pesakovicet *al.*,(2013) and Rayees *et al.*,(2013) in strawberry.

Investigation revealed that the biofertilizers and organic manures significantly influence the plant reproductive growth (fruit set), fruit quality (TSS, ascorbic acid, acidity, total sugar), physiochemical properties (specific gravity and juice content), and productivity (fruit length, fruit weight, fruit diameter, fruit volume) of strawberry fruit var. Camarosa.

Table02:Pearson correlation matrix of Camarosa strawberry affected by soil amendments

	Productivity	Fruit set	Fruit length	Fruit diameter	Fruit weight	Fruit volume	Specific gravity	Juice content	TSS	Acidity	Ascorbic acid	Total sugar
Productivity	1.000	0.674*	0.860**	0.823*	0.951**	0.961**	0.552 ^{NS}	0.961*	0.920**	0.961*	0.870**	0.927**
Fruit set	0.674*	1.000	0.652*	0.681*	0.730*	0.743*	0.538 ^{NS}	0.623 ^{NS}	0.583 ^{NS}	0.641*	0.913**	0.773**
Fruit length	0.860**	0.652*	1.000	0.876*	0.881**	0.839**	0.748*	0.891*	0.887**	0.902*	0.783**	0.890**
Fruit diameter	0.823**	0.681*	0.876**	1.000	0.886**	0.845**	0.772**	0.869*	0.817**	0.860*	0.790**	0.791**
Fruit weight	0.951**	0.730*	0.881**	0.886*	1.000	0.989**	0.690*	0.942*	0.921**	0.959*	0.871**	0.924**

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Fruit volume	0.961**	0.743*	0.839**	0.845*	0.989**	1.000	0.584 ^{NS}	0.940*	0.901**	0.955*	0.880**	0.936**
Specific gravity	0.552 ^{NS}	0.538 ^{NS}	0.748*	0.772*	0.690*	0.584 ^{NS}	1.000	0.608 ^{NS}	0.616 ^{NS}	0.627 ^{NS}	0.600 ^{NS}	0.557 ^{NS}
Juice content	0.961**	0.623 ^{NS}	0.891**	0.869*	0.942**	0.940**	0.608 ^{NS}	1.000	0.869**	0.996*	0.792**	0.919**
TSS	0.920**	0.583 ^{NS}	0.887**	0.817*	0.921**	0.901**	0.616 ^{NS}	0.869*	1.000	0.882*	0.792**	0.875**
Acidity	-0.961**	0.641*	0.902**	0.860*	0.959**	-0.955**	-0.627 ^{NS}	0.996*	0.882**	1.000	0.804**	0.938**
Ascorbic acid	0.870**	0.913*	0.783**	0.790*	0.871**	0.880**	0.600 ^{NS}	0.792*	0.792**	0.804*	1.000	0.900**
Total sugar	0.927**	0.773*	0.890**	0.791*	0.924**	0.936**	0.557 ^{NS}	0.919*	0.875**	0.938*	0.900**	1.000

The significance levels are indicated using symbols like "*", "**," and "NS":

"*" represents a moderate positive correlation significant at the 0.05 level.

"**" represents a strong positive correlation significant at the 0.01 level.

"NS" (not significant) indicates that the correlation is not statistically significant.

"-**" represents that the correlation is negatively significant.

Table 2 provides the correlation coefficients between pairs of variables in the dataset. The Pearson correlation coefficient measures the strength and direction of a linear relationship between two variables. The value of the coefficient ranges from -1 to 1. The variable "productivity" has a strong positive correlation with variables like fruit length, fruit diameter, fruit weight, fruit volume, juice content, TSS, ascorbic acid, and total sugar. "Fruit set" has a moderate positive correlation with variables like productivity, fruit length, fruit diameter, fruit weight, and fruit volume, and a strong positive correlation with ascorbic acid and total sugar. Variables like fruit length, fruit diameter, fruit weight, fruit volume, and juice content have strong positive correlations with many other variables, indicating that they tend to increase together. Moreover, "specific gravity" is not correlated with many variables except fruit length, fruit diameter, and fruit weight.

Juice content and TSS have strong positive correlations with other variables but have no correlation with specific gravity or fruit set. Ascorbic acid and total sugar have strong positive correlations with other variables but have no correlation with specific gravity. "Acidity" has strong negative correlations with other variables, suggesting that as acidity increases, other attributes tend to decrease.

Conclusion

From the above findings, it is evident that the simultaneous utilization of biofertilizers and organic manures yields superior quality fruits. This outcome is attributed to the heightened nutrient availability within the soil, subsequently benefiting the host plant. Furthermore, the positive influence extends to the reproductive growth and overall productivity of the plant. Consequently, this combined approach holds significant promise for fostering the cultivation of high-quality strawberries in a sustainable manner. Embracing this technique could also contribute to the advancement of sustainable agricultural practices, aligning with the current imperative for organic farming principles. As demand for sustainable food production continues to rise, this integrated strategy offers a potential solution for meeting these heightened expectations while ensuring environmentally conscious and efficient agricultural operations.

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Author Contribution: JT and AT: investigations, data collection, visualization and editing. AK: data curation, data analysing, conceptualization, methodology, supervision, writing-original draft, review and editing.

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Data Availability As this article incorporates entirely original data, data sharing is not relevant to its content.

Declarations

Conflict of Interests: The authors declare no competing interests.

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