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Influence of Biostimulants and Organic mulch on soil Microbial population in Strawberry (F. × ananassa Dutch.) cv. Katrain Sweet

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Abstract

To investigate the impact of biostimulants and organic mulch on soil microbial population of strawberry cv. Katrain Sweet, an experiment was carried out in the Biocontrol lab, Chandra Shekhar Azad University of Agriculture and Technology, Kanpur, during two subsequent years *i.e.,* 2022-2023 and 2023-2024. The results showed substantial enhancements in microbial growth of both fungus and bacteria with the use of *Azotobacter*, *Trichoderma* and PSB applied in combined with organic mulch, particularly in treatment *T_{12}-Azotobacter (*8g/plant) + *PSB (*8g/plant) + Trichoderma *(*6g/plant) + dried leaves. The findings underscore the potential for the use of biostimulants and organic mulches to enhance soil microbial growth in strawberry production, offering a sustainable approach for improving yield and plant growth.

Keywords: biostimulants; *azotobacter*; *trichoderma;* dried leaves

Introduction

The strawberry (*Fragaria* × *ananassa*) belongs to the family Rosaceae. It is a hybrid species, originating from a cross between the wild species *Fragaria virginiana* and *Fragaria chiloensis*. Strawberries are classified as aggregate accessory fruits which means that the fleshy part is derived not from the plant's ovaries but from the receptacle that holds the ovaries. The tiny "seeds" on the surfaces called as achenes which are true fruits and contains a single seed **Rousseau (2009), Tripathi (2010)**. Botanically, strawberries are herbaceous perennials with a low-growing habit. They spread via stolon's (runners) which are horizontal above-ground stems that produce new plants. The leaves are trifoliate typically with serrated edges and arise from a central crown. White or pink five-petal flowers emerges which leads to fruit development. Strawberries thrive in well-drained soils with a pH of 5.5 to 6.5, requiring full sunlight for optimal growth and fruit production **Kafkas (2023), Tripathi (2016)**.

Azotobacter is a free-living nitrogen-fixing bacterium that enhances soil fertility by converting atmospheric nitrogen into a form that plants can absorb, such as ammonium. It promotes plant growth by producing growth hormones like auxins, cytokinins and gibberellins **(Calvo, 2014)**. This nitrogen fixation helps reduce the need for chemical fertilizers and improves soil structure by enhancing organic matter. Azotobacter contributes to an increase in nitrogen content in the soil which promotes the growth of beneficial microbes. By producing growth hormones and biologically available nitrogen, Azotobacter fosters the proliferation of symbiotic and free-living microbes that thrive in nitrogen-rich environments. This bacterium also produces antimicrobial substances that can suppress harmful microbial populations, thus maintaining a balance in the soil's microbial ecosystem **(Ayangbenro, 2022)**.

Phosphate-Solubilizing Bacteria (PSB) help in solubilizing insoluble forms of phosphorus in the soil while making its availability to plants. By secreting organic acids like gluconic acid they dissolve bound phosphates which increases phosphorus uptake **(Bashan, 2014)**. This reduces the need for synthetic phosphate fertilizers, enhances root growth and supports the development of strong with healthy plants. By solubilizing phosphate, these bacteria not only aid plant nutrition but also stimulate the growth of other microorganisms that depend on phosphorus for their metabolism. This results in an increase in microbial diversity, as more species can thrive in the enriched soil environment. PSBs also engage in mutualistic relationships with other microbes, further enhancing microbial biodiversity **(Raghothama. 1999)**.

Trichoderma is a beneficial fungus that acts as a biocontrol agent against soil-borne pathogens. It competes for space and nutrients, inhibits harmful fungi through parasitism and induces plant resistance. Trichoderma also promotes root growth, enhances nutrient uptake and improves soil structure by increasing organic matter breakdown **(Manzar, 2023)**. Trichoderma acts as a natural biocontrol agent, suppressing pathogenic fungi and creating a favourable environment for beneficial microbes. By competing for nutrients and space, Trichoderma reduces the population of harmful pathogens while promoting beneficial fungi and bacteria. Its ability to degrade organic matter enriches the soil with nutrients, fostering the growth of various microorganisms. The introduction of Trichoderma can lead to an increase in microbial biomass and activity and supporting overall soil health **(Noumavo, 2016)**.

Material and methods

The crop research was carried out in the Garden, Department of Fruit Science, Chandra Shekhar Azad University of Agriculture and Technology, Kanpur (U.P.), during two subsequent years *i.e.*, 2022-2023 and 2023-2024. The Kanpur district is located at an elevation of 135 meters above sea level. It is situated in a subtropical zone with coordinates between 25.26° & 26.58° North latitude & 79.31° and 80.34° Eastern longitude. Several treatments were employed for optimal results, it is recommended to utilize paddy straw and dried leaves as organic mulch and application of 5-8 g of *Azotobacter*, 5-8 g of *Trichoderma* and 5-8 g of PSB per plant. The soil samples were collected from rhizosphere of the strawberry and subjected to analysis of soil microbes (bacteria and fungi) after completion of strawberry cultivation at Bio control lab at same university.

The following method was adopted for preparation of culture media for growing of bacterial and fungal growth. The relevant information in Table-1

Potato Dextrose Agar (PDA) medium for fungal growth	Plate Count Agar (PCA) media for bacterial growth
Extract from peeled potatoes (200 g)	Tryptone (5 g)
Dextrose (20 g)	Yeast extract (2.5 g)
Agar- agar (20 g)	Dextrose (1 g)
Distilled water (1000 ml)	Agar-agar (15 g)
-	Distilled water (1000 ml)

Table-1: Preparation of Medias for growth of microbes.

50 g soil samples were collected from rhizosphere (closed to roots) of strawberry plants and collected in sterile polyethylene bags, which were transported to the laboratory and stored at 4 °C until use. The serial dilation techniques were adopted for further investigation **(Johnson and Curl, 1972)**.

Ten-gram soil sample from well pulverized and air-dried soil was added into 90 ml sterile water in a flask to make 1:10 dilution (10^{-1}). The mixture was vigorously shaken on a magnetic shaker for 20-30 minutes to obtain uniform suspension. One ml of suspension from flask was transferred into a test tube containing 9 ml sterile water under aseptic condition to make 1:100 (10^{-2}) dilution. Further dilution 10^{-3} was made by pipetting 1 ml suspension into additional water as prepared above.

One ml each liquid of 10^{-4} dilution (for fungus) and 10^{-6} dilution (for bacteria) were transferred into sterile Petri plates, which was previously poured by 15 ml sterile PDA medium (fungus) and PCA (bacteria) and spread uniformly under laminar flow. The plates were sealed properly with para film. The plates were kept upright (fungus) and inverted (bacteria) in BOD Incubator. After 72-96 hours (PDA plates) and 48-72 hours (PCA plates) incubation of microbe population were counted with the help of Colony Counter.

Result and discussion

Bacterial growth population

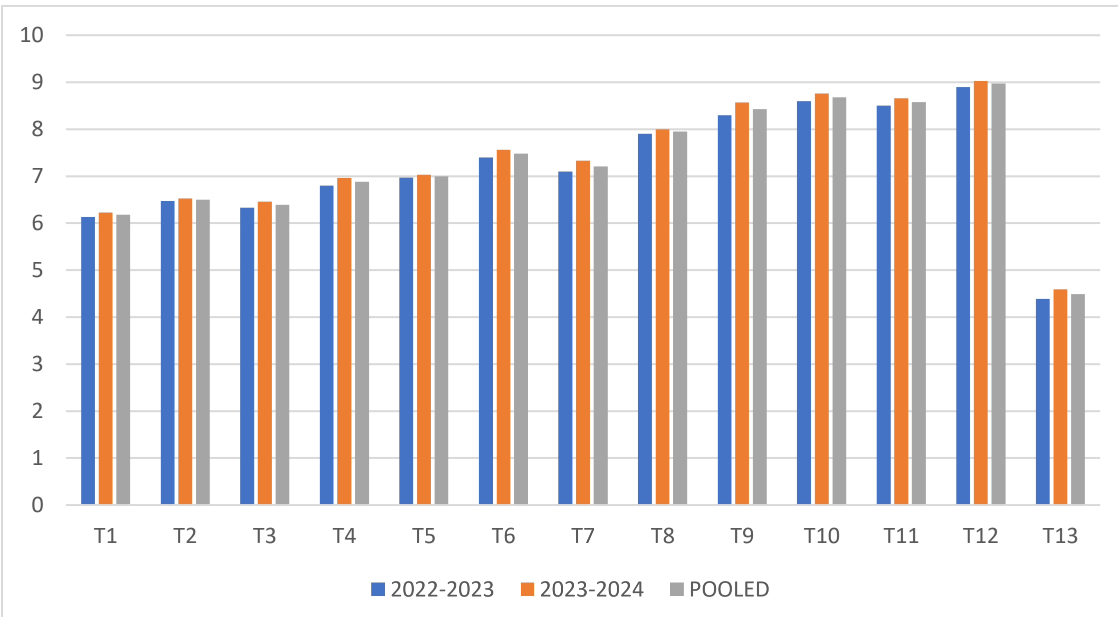
Both years data revealed that combination of T_{12} (*Azotobacter* (8g/ plant) + PSB (8g/ plant) + *Trichoderma* (6g/ plant) + dried leaves) resulted in maximum bacterial population count at 8.97×10^6 cfu g^{-1} of soil, indicating its superior effect on bacterial growth. In contrast, the control treatment (T_{13}) recorded the lowest bacterial count of 4.49×10^6 cfu g^{-1} of soil (Table-2). Effect of bio-fertilizer and organic mulch on bacterial population in soil was found statistically significant. This is certainly because the *Azotobacter* and PSB helps in multiplication of bacterial population, which is necessary for optimum growth of strawberry plants **Kundu (2024), Gargi N (2007)** and **Kher R (2010)**.

Fungal growth population

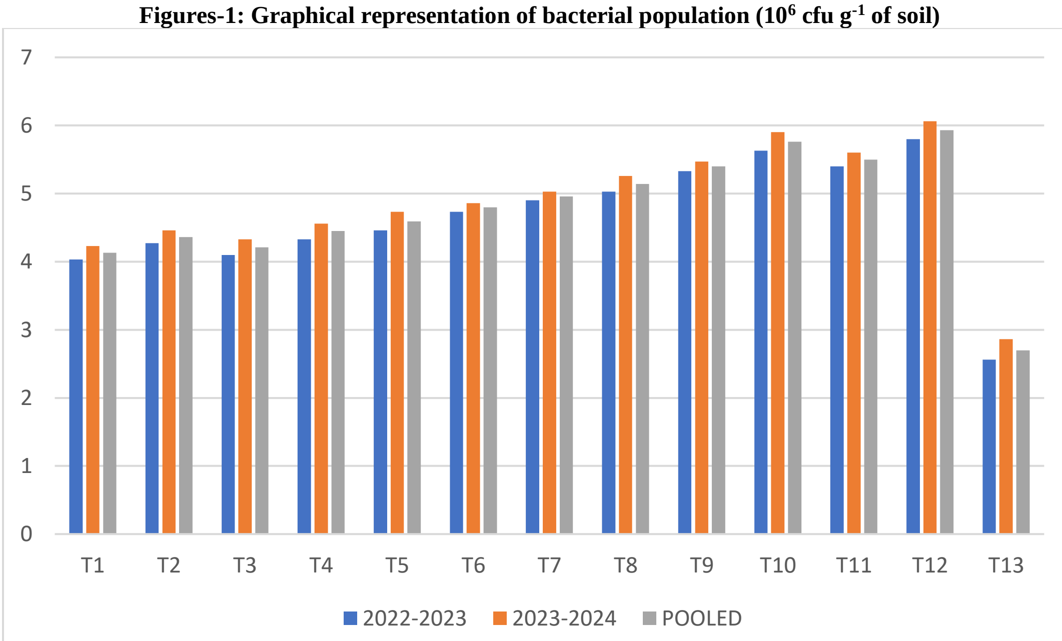
Treatments involving *Azotobacter* (8g/plant) + PSB (8g/plant) + *Trichoderma* (6g/plant) + dried leaves (T_{12}) produced highest fungal population with mean of 5.96×10^4 cfu g^{-1} of soil. These treatments significantly outperformed the control with fungal population at only 2.70×10^4 cfu g^{-1} of soil (Table-2). Effect of bio-fertilizer and organic mulch on fungal population in soil was found statistically significant. Trichoderma proves to be effective in increasing the fungal population and reducing the chances of possible pathogen infestation in strawberry **Chauhan (2010), Derkowska (2015)** and **Garg (2011).**

Treatments	Bacterial population (10^6 cfu g^{-1} of soil)			Fungal population (10^4 cfu g^{-1} of soil)		
	2022-23	**2023-24**	**Pooled**	**2022-23**	**2023-24**	**Pooled**
T_1-Azotobacter **(5g/plant) + Trichoderma (5g/plant) + paddy straw**	6.13	6.23	6.18	4.03	4.23	4.13
T_2-Azotobacter **(8g/plant) + Trichoderma (6g/ plant) + paddy straw**	6.47	6.53	6.50	4.27	4.46	4.36
T_3-Azotobacter **(5g/plant) + Trichoderma (5g/plant) + dried leaves**	6.33	6.46	6.39	4.10	4.33	4.21
T_4-Azotobacter **(8g/plant) + Trichoderma (6g/plant) + dried leaves**	6.80	6.96	6.88	4.33	4.56	4.45
T_5-Azotobacter **(5g/plant) + *PSB* (5g/plant) + *paddy straw***	6.97	7.03	7.00	4.46	4.73	4.59
T_6-Azotobacter **(8g/plant) + *PSB* (8g/plant) + *paddy straw***	7.40	7.56	7.48	4.73	4.86	4.80

T_{7}-Azotobacter* (5g/plant) + *PSB* (5g/plant) + *dried leaves	7.10	7.33	7.21	4.90	5.03	4.96
***T_{8}-Azotobacter* (8g/plant) + *PSB* (8g/plant) + dried leaves**	7.90	8.00	7.95	5.03	5.26	5.14
***T_{9}-Azotobacter* (5g/plant) + *PSB* (5g/plant) + Trichoderma (5g/plant) + paddy straw**	8.30	8.57	8.43	5.33	5.47	5.40
***T_{10}-Azotobacter* (8g/plant) + *PSB*(8g/plant) + Trichoderma (6g/plant) + paddy straw**	8.60	8.76	8.68	5.63	5.90	5.76
***T_{11}-Azotobacter* (5g/plant) + *PSB* (5g/plant) + Trichoderma (5g/plant) + dried leaves**	8.50	8.66	8.58	5.40	5.60	5.50
***T_{12}-Azotobacter* (8g/plant) + *PSB* (8g/plant) + Trichoderma (6g/plant) + dried leaves**	8.90	9.03	8.97	5.80	6.06	5.93
T_{13}-Control	4.39	4.59	4.49	2.56	2.86	2.70
CD at 5% level	0.32	0.49	0.15	0.33	0.47	0.28
CV%	0.94	1.43	0.45	0.96	1.37	0.87



Figures-1: Graphical representation of bacterial population (10^6 cfu g^{-1} of soil)



Figures-2: Graphical representation of fungal population (10^4 cfu g^{-1} of soil)

Conclusion

The combination of *Azotobacter* (8g/plant) + PSB (8g/plant) + *Trichoderma* (6g/plant) with dried leaves shown significant effect on soil microbial population with effective growth in fungus and bacteria. Based on these results, the integration of biostimulants and organic mulches into strawberry cultivation practices can result in sustainable and efficient production which is advantageous to both the environment and cultivators in the plains of North India.

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