Response of organic manure and Azotobacter on quality and leaf nutrient status of strawberry (*Fragaria* x *ananassa* Duch.) cv. Winter Dawn

ABSTRACT

The ~~Field~~ field experiment was conducted during two subsequent years, *i.e.*, 2017-18 and 2018-19 at the Department of Horticulture, Hisar with aim to study the effect of organic manures and *Azotobacter* on quality and leaf nutrient status of strawberry cv. Winter Dawn under hi-tech greenhouse conditions of Haryana. In this experiment, organic manures in combination with biofertilizers comprised of nine treatments, *viz.*, T_1: Sand: FYM (3:1), T_2: Sand: FYM (2:1), T_3: Sand: Vermicompost (3:1), T_4: Sand: Vermicompost (2:1), T_5: Sand: FYM (3:1) + *Azotobacter*, T_6: Sand: FYM (2:1) + *Azotobacter*, T_7: Sand: Vermicompost (3:1) + *Azotobacter*, T_8: Sand: Vermicompost (2:1) + *Azotobacter* and T_9: Control (Sand); were tested as potting media in complete randomized block design. Maximum TSS, ascorbic acid, anthocyanin content and minimum acidity content and leaf with maximum N, P and K content were observed under sand: vermicompost (2:1) + *Azotobacter* in comparison to the other treatments. The results of this experiment revealed that, the combination of sand: vermicompost (2:1) with Azotobacter showed significant influence on leaf nutrient content of strawberry and produced better results with respect to quality of strawberry fruits.

Key words: Strawberry. organic manure. *Azotobacter*. Quality.

1. INTRODUCTION

The modern cultivated strawberry (*Fragaria* x *ananassa* Duch.), is an inter-specific hybrid of two new world species, *Fragaria virginiana* Duch., a wild strawberry native to Virginia and *Fragaria chilonensis* (L.) Duch., a South American strawberry, and belongs to the family Rosaceae. Fruits are popular for their distinctive succulent nature, bright red colour, juicy texture, aroma and are a rich source of vitamins, minerals and phenolics. Strawberry plants having shallow root system need ~~i~~effective and balanced nutrient management, thus, nutrient status of soil is most important factor affecting the production of strawberry. The modern-day intensive crop cultivation involves bulk application of inorganic fertilizers, which are not only in short supply but also costly. They also pollute the environment, soil and water. Farmyard manure and vermicompost are the main organic components for horticultural crop production [2,3].

Biofertilization is the new concept that is being adopted in numerous agricultural and horticultural crop production systems, reducing cost of production and minimizing environmental pollution [4]. Nitrogen fixing bacteria (*Azosprilllum*, *Azotobactor*), phosphate solubilizers (*Pseudomonas*, *Bacillus*) and phosphate absorbers (*Mycorrhizal* fungi) are the most important biofertilizers applied for horticultural crop cultivation [5]. *Azotobactor* is a free-living heterotrophic nitrogen fixing bacteria that enhances plant growth by stimulating growth promoting substances such as auxins, gibberellins and vitamins *etc.* [6,7]. Vermicompost supplies all essential macro elements such as N, P, K, Ca, Mg and microelements such as Fe, Zn, Mo and Co to fulfil plant requirements during the crop growth period . The application of bio-fertilizer along with organic or inorganic fertilizers significantly increases fertilizer use efficiency, nutrient uptake, growth and yield of strawberry [8].

Comment [JA1]: This title ought to be paraphrased to either

1.Effect of Organic Manure and Azotobacter on quality and leaf nutrient status of strawberries (*Fragaria* x *ananassa* Duch.) cv. Winter Dawn

2.Quality and leaf nutrient status of strawberry (*Fragaria* x *ananassa* Duch.) cv. Winter Dawn grown under organic manure and Azotobacter

In its present form, it is not sounding academic

Comment [JA2]: This study period is a little confusing. May be the authors need to include the months

Comment [JA3]: Found in which country?

Comment [JA4]: Found in which country?

Comment [JA5]: Arrange in alphabetical order.

Also, the study site is a keyword. The authors should include it in the set of new words.

Comment [JA6]: This should have been presented first. Equally, the authors should labour to acknowledge the source from which they extracted this information.

I also suggest that the authors mention the common fruits consumed by man and include strawberries in the list . They should include sources of information cited.

Then , the authors should cascade down to a brief history of strawberries and the chief ...

Comment [JA7]: There is need for empirical studies on the same crop in india in relation to effects on the levels of soil fertility before the application of inorganic fertilizers is brought out.

Comment [JA8]: Where is the source of this information?

Comment [JA9]: What is farm yard manure? What is vermicompost? How are they made? What is the capacity of India to produce these? What are the enabling factors? The authors should use the latest ...

Comment [JA10]: What is Bio fertilization? Why is it being adopted in horticulture system? Where is it commonly applied? What does it involve? What are the components o...

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Hence, the application of vermicompost and *Azotobacter* should help to produce quality berries with higher yields. ~~Keeping the above facts in view, a field trial was conducted~~

In India, strawberry cultivation is primarily confined to temperate zones of Uttarakhand, Himachal Pradesh, Jammu and Kashmir, hills of West Bengal (Darjeeling), Tamil Nadu (Ooty) and the tropical and subtropical zones of Haryana, Uttar Pradesh, Punjab, Maharashtra (Mahabaleshwar) and Karnataka (Bengaluru) with an area of 1000 hectare and production of 5000 tonne [1].

Keeping the above facts in view, a field trial was conducted.to ……???? And so what was the problem be addressed by this study? The authors should ensure that the aim of the study and the problem to be addressed are clearly shown at this point before delving into the materials and methods section. ~~.~~

2. MATERIALS AND METHODS

The present investigations were carried out at Hitech- greenhouse, CCS Haryana Agricultural University, Hisar, (Haryana) in which country? during the year 2017-18 and 2018-19, to study the effect of organic manures with biofertilizers on quality and leaf NPK status of strawberry cv. Winter Dawn. Uniform runners were selected for planting and single healthy uniform runners were planted in each pot after treating with copper oxychloride (0.1%) for 10 ~~minutes~~minutes. The planting was done in the 2nd week of October in both consecutive years (2017-18 and 2018-19). There ~~are nine~~were nine treatments, *viz*., T_1: Sand: FYM (3:1), T_2: Sand: FYM (2:1), T_3: Sand: Vermicompost (3:1), T_4: Sand: Vermicompost (2:1), T_5: Sand: FYM (3:1) + *Azotobacter*, T_6: Sand: FYM (2:1) + *Azotobacter*, T_7: Sand: Vermicompost (3:1) + *Azotobacter*. T_8: Sand: Vermicompost (2:1) + *Azotobacter* and T_9: Control (Sand), replicated five times in complete randomized block design. Each twelve inches' pot was filled with sand, FYM and vermicompost; volume by volume on basis of treatment imposition. *Azotobacter* were inoculated @10 ml per plant in the form of liquid at 20 and 40 days after transplanting in the pots.

The recommended dose of fertilizers (NPK 1.95: 0.8:2.75 g plant^{-1}) was applied in the form of water-soluble fertilizers (Urea, 19:19:19 and KNO_3). Fertigation was done at weekly intervals after ~~a week after~~ trans planting. Strawberry plants were subjected to uniform application of recommended dose of fertilizers, plant protection measures and other cultural practices. Observations on various quality parameters were recorded by using standard methods. Twenty fruits from each treatment were randomly selected to record the data on quality parameters. TSS was measured with the help of ERMA hand refractrometer (0-32 °Bx), Titratable acidity (%), anthocyanin and vitamin C (mg/100 gm) were estimated by using the method suggested by [9]. Total N was determined by micro-Kjeldahl method [10]. Diacid mixture was used for wet digestion of samples to determine the P and K. Phosphorus was determined by Vanadomolybo-phosphoric acid yellow colour method and potassium content in leaf samples was estimated by flame photometric method [11]. The data was ~~analyzed~~analysed with the help of a window-based computer package OPSTAT [12], to calculate standard error of means SE(m), standard error of difference in mean SE(d), and critical difference between treatments mean CD.

3. RESULTS AND DISCUSSION

Comment [JA12]: The contextual aspect of India and her production potential of strawberries should appear here followed by the problem of the study and the research objective pursued by the authors.

The authors are requested to include a table showing the trend of strawberry production in these zones. This should be accessed from Bureau of Statistics or Indian Department of Agricultural Production.

Comment [JA13]: Why did the experiment take two years? Some scientific explanation needed here.

Comment [JA14]: What criteria was followed to ensure uniformity??

Comment [JA15]: Where were these sourced from and why this area?

Comment [JA16]: Which material was the pot made of and why?

Comment [JA17]: What was the essence of treating the pots with copper oxychloride?

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Comment [JA19]: What equipment was used in transplanting and why was it used?

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3.1 Total soluble solids (^{0}Brix): TSS of strawberry fruits was significantly affected by the application of different ratios of organic manures and *Azotobacter* with respect to various treatments as revealed ~~in~~ by Table 1. The maximum TSS were observed under Sand: Vermicompost (2:1) + *Azotobacter* (T_8), whereas the minimum TSS was recorded from the control (T_9) plants during both years of investigation. An increase in TSS with plant growth promoting bacteria might be due to the quick metabolic transformation of starch and pectin into soluble compounds and rapid translocation of sugars from leaves to the developing fruits [13]. The results of present study are in harmony with the findings of [14] who recorded maximum TSS in fruits harvested from the plants grown under organic manures with biofertilizers, which might be due to the reason that biofertilizers in combination with *Azotobacter* increased the accumulation of carbohydrates and metabolites which were~~h~~ converted into disaccharides leading to higher TSS in strawberry fruits. Similar results were also obtained by authors [15~~,16,17,18~~, 16, 17, 18] in strawberry.

3.2 Titratable acidity (%): The effect of various treatments on acidity of strawberry fruits is shown in Table 1. The minimum titratable acidity was recorded in sand: vermicompost (2:1) + *Azotobacter* (T_8), which was at~~on~~ par with the application of sand: vermicompost (3:1) + *Azotobacter* (T_7) and maximum was recorded in T9 (Control) over the other treatments. The reduction in titratable acidity may be attributed to conversion of organic acids and photosynthates into sugar during fruit ripening by applying biofertilizers [19]. These findings are in close conformity with the results of [20] who observed that the reduction in titratable acidity may also be due to utilization of acids as a substrate for respiration during the ripening and neutralization of organic acids due to potassium in tissues. Similar findings were also reported [21, 22] in strawberry.

3.3 Ascorbic acid (mg/ 100 ml): The maximum ascorbic acid content was observed under sand: vermicompost (2:1) + *Azotobacter* (T_8), whereas the minimum was ~~recorded~~recorded from the control (T_9) plants during both years of investigation as shown in Table 1. The increase in ascorbic acid content might be due to the increased efficiency of microbial inoculants to fix atmospheric nitrogen, increase in availability of nitrogen and excretion of growth promoting hormones which accelerates the physiological process like carbohydrates synthesis, etc. An increase in ascorbic acid content with *Azotobacter* and vermicompost application might be due to the quick metabolic transformation of starch and pectin into soluble compounds and rapid translocation of sugars from leaves to the developing fruits [14]. Similar results were found by authors [23, 24] in strawberry.

3.4 Anthocyanin (mg/100 mg): Anthocyanin significantly increased by the application of combination of organic manures and bio-fertilizer over the control. The maximum anthocyanin contents were observed under sand: vermicompost (2:1) + *Azotobacter* (T_8), whereas the minimum was recorded from the control (T_9) plants during both years of investigation as shown in Table 1. These results may be due to the synergistic effect of macronutrients supplied through organic and bio-fertilizer which improved the vegetative characteristics of the plants thereby affecting ~~and affected~~ the synthesis of chlorophyll that enhanced the process of photosynthesis and the assimilation of carbon dioxide which led to increased fruit quality [25].

Discussion of the results is missing at this point…….AUTHOR SHOULD INCLUDE IT.

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Comment [JA33]: This is a major point of discussion of the results by the authors themselves by appealing to scientific principles. So they SHOULD NOT attribute their discussion to another study since the findings here are their voice. This is catered for by the following sentence

Comment [JA34]: This is a major point of discussion of the results by the authors themselves by appealing to scientific principles. So they SHOULD NOT attribute their discussion to another study since the findings here are their voice. This is catered for by the following sentence

Table 1. Effect of organic manures in combination with biofertilizers on quality parameters of strawberry

Treatments	TSS (^{0}Brix)		Acidity (%)		Ascorbic acid (mg/100 ml)		Anthocyanin (mg/100 ml)	
	2018	2019	2018	2019	2018	2019	2018	2109
T_1: Sand: FYM (3:1)	6.58	6.70	0.82	0.82	42.48	43.04	34.48	35.65
T_2: Sand: FYM (2:1)	6.64	6.79	0.82	0.80	42.69	43.38	35.82	36.90
T_3: Sand: Vermicompost (3:1)	6.95	7.10	0.79	0.78	47.11	47.66	39.33	40.54
T_4: Sand: Vermicompost (2:1)	7.05	7.11	0.78	0.77	47.83	48.32	40.62	41.62
T_5: Sand: FYM (3:1) +*Azotobactor*	7.08	7.18	0.77	0.76	45.98	47.11	39.75	40.66
T_6: Sand: FYM (2:1) +A*zotobactor*	7.13	7.22	0.76	0.74	46.11	47.32	41.15	42.02
T_7: Sand: Vermicompost (3:1) +*Azotobactor*	7.45	7.56	0.73	0.71	50.22	51.39	43.52	44.15
T_8: Sand: Vermicompost (2:1) +*Azotobacto*r	7.59	7.73	0.70	0.68	51.01	52.15	45.27	45.92
T_9: Control: (Sand only)	6.03	6.08	0.83	0.82	40.17	40.70	31.24	32.07
CD at 5%	0.62	0.60	0.05	0.07	2.43	2.28	1.99	1.79

3.5 N, P and K status of leaf: The leaf nutrient content was influenced significantly by the organic manure in combination with biofertilizer (Table 2). The maximum leaf N, P and K content were recorded under (T_8) sand: vermicompost (2:1) + *Azotobacter* treatment followed by T_7, T_6 and T_5 and minimum in the control (T_9). Plants fertilized with vermicompost have shown greater ability to assimilate essential macronutrients as also observed by other researchers [26]. The higher uptake of nitrogen and phosphorous may be due to improved symbiotic N_2 fixation and also due to improved phosphatase activity, thereby improving phosphorous mobilization and uptake from the root zone. The increase in uptake of nutrients may be due to extra amount of nutrients supplied by the organic fertilizers and provision of conducive physical environment which helps in better root growth and absorption of nutrients from the soil [27]. The results of present study are in line with the findings of [4] who observed that maximum contents of nitrogen, phosphorus and potassium were observed in guava with recommended dose of fertilizers along with vermicompost in guava cv. Sardar; and [28] who recorded higher leaf macro nutrient content in strawberry plants fertilized with *Azotobactor* in combination with organic fertilizers in comparison to control.

Table 2. Effect of organic manures in combination with biofertilizers on leaf N, P and K content of strawberry

Treatments	N (%)		P (%)		K (%)	
	2018	2019	2018	2019	2018	2019
T_1: Sand: FYM (3:1)	2.54	2.61	0.34	0.41	1.23	1.39

Comment [JA35]: We have not lived this year yet.....So correct to 2019

Comment [JA36]: Content best fits here before the table

Comment [JA37]: This is a major point of discussion of the results by the authors themselves by appealing to scientific principles. So they SHOULD NOT attribute their discussion to another study since the findings here are their voice. This is catered for by the following sentence

Comment [JA38]: Mention the name of the author here

T_2: Sand: FYM (2:1)	2.65	2.72	0.39	0.46	1.38	1.46
T_3: Sand: Vermicompost (3:1)	2.75	2.78	0.41	0.50	1.41	1.51
T_4: Sand: Vermicompost (2:1)	2.81	2.85	0.48	0.57	1.53	1.59
T_5: Sand: FYM (3:1) +*Azotobactor*	2.70	2.74	0.40	0.51	1.40	1.52
T_6: Sand: FYM (2:1) +A*zotobactor*	2.77	2.86	0.49	0.57	1.51	1.63
T_7: Sand: Vermicompost (3:1) +*Azotobactor*	2.96	3.01	0.50	0.60	1.56	1.62
T_8: Sand: Vermicompost (2:1) +*Azotobacto*r	3.06	3.08	0.60	0.69	1.69	1.71
T_9: Control: (Sand only)	1.98	2.04	0.24	0.31	1.05	1.13
CD at 5%	0.09	0.07	0.06	0.05	0.12	0.07

~~**3.5 N, P and K status of leaf:** The leaf nutrient content was influenced significantly by the organic manure in combination with biofertilizer (Table 2). The maximum leaf N, P and K content were recorded under (T_8) sand: vermicompost (2:1) + *Azotobacter* treatment followed by T_7, T_6 and T_5 and minimum in the control (T_9). Plants fertilized with vermicompost have shown greater ability to assimilate essential macronutrients as also observed by other researchers [26]. The higher uptake of nitrogen and phosphorous may be due to improved symbiotic N_2 fixation and also due to improved phosphatase activity, thereby improving phosphorous mobilization and uptake from the root zone. The increase in uptake of nutrients may be due to extra amount of nutrients supplied by the organic fertilizers and provision of conducive physical environment which helps in better root growth and absorption of nutrients from the soil [27]. The results of present study are in line with the findings of [4] who observed that maximum contents of nitrogen, phosphorus and potassium were observed in guava with recommended dose of fertilizers along with vermicompost in guava cv. Sardar; and [28] who recorded higher leaf macro nutrient content in strawberry plants fertilized with *Azotobactor* in combination with organic fertilizers in comparison to control.~~

4. Conclusion

From the results, it ~~may be revealed~~is ~~that~~concluded that quality strawberry plants ~~raised~~are raised with organic manures ~~in combination with~~ fortified by biofertilizers ~~recorded better quality parameters~~. This was shown by the a~~A~~pplication of Sand: vermicompost (2:1) along with the combination of *Azotobacto*r which showed significantly increased quality parameters as well as maximum leaf NPK content~~,~~ compared to ~~then~~ other treatments. Then what is the most plausible recommendation arising from your study that can inform farmers, decision makers, organizations biased on promoting strawberries and other fruits as well as firms dealing in production of vermicompost and bio fertilizers????

References

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Comment [JA39]: Observe the citation and referencing protocols of the journal as you add more references following the recommendations and suggestions made by the reviewer.

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Comment [JA40]: What is the difference between this citation and Number 6? There is some disorder here that MUST be thoroughly addressed

17. Mahavir S, Devi SK, Sandeep S, Mahendra B. Effect of integrated nutrient management on growth, yield and quality traits of strawberry (*Fragaria* x *ananassa* Duch.) cv. Chandler. Journal of Pharmcognacy and Photochemistry. 2018;1: 712-715.
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Comment [JA41]: There is inconsistence here. Look at (2012) in 26 and 2016 in 27. The authors are not attending to detail in punctuating the citations in the list of references. We must observe uniformity.

Check up again, the same digression is appearing in number 15, 22, 12