Growth and Quality proficency of onion (*Allium cepa* L.) by application of biofertilizer, GA_3 and Humic acid

ABSTRACT

During the Rabi season in 2022-2023, a comprehensive trial was carried out at the Department of Horticulture, Jawaharlal Nehru Krishi Vishwa Vidyalaya. Employing a Randomized Completely Block Design (RCBD), the research assessed twelve different interventions, consisting of biofertilizers, GA_3, humic acid and a reference group. Notably, the combined application of Azotobacter, Azospirillum, GA_3 and humic acid (T10) showcased substantial improvements in several growth parameters. These encompassed plant height (62.23 cm), number of leaves (10.18), length of leaves (54.08 cm), leaf area (624.65 cm²), leaf area index (4.16) and leaf diameter (8.14 mm). Additionally, this treatment recorded the highest Total Soluble Solids (TSS) at 12.37 °Brix. Moreover, the inclusion of sulphur and humic acid significantly affected the pyruvic acid content, with T10 recorded the highest pyruvate values (5.28 µmol/g). Conversely, the reference group (T12) only recorded slight improvements, emphasizing the noteworthy impact of the interventions on both the growth and qualitative characteristics of the onions.

Keywords: Biofertilizers; GA_3 and humic acid; onion growth parameters; total soluble solids (TSS); pyruvic acid content.

INTRODUCTION

The onion, referred to as the "Ruler of the Kitchen," is a biennial herb that has become a fundamental part of cooking customs worldwide. Originating from Central Asia and the Mediterranean area, it has transformed into a vital seasoning crop. It is widely farmed in different parts of India throughout both the kharif and rabi seasons, indicating its importance in the farming landscape.

Onions (*Allium cepa* L.), which are highly prized for their high nutritional content, are widely produced for year-round eating (Roldan et al., 2008 and Sekara et al., 2017) [12,14]. Biofertilizers, which are microorganism inoculants used in seed inoculation, convert inaccessible nutrients into accessible forms, hence improving crop growth (Aswani et al., 2005 and Fawy et al., 2016) [1,6]. According to research by Wu et al. (2005) and Kumar et al. (2019) [16,10], these biofertilizers boost both the quality and quantity of onions.

The use of bio-fertilizers, like Azotobacter and Azospirillum, provides a cost-effective and effective approach to enhance crop growth by boosting nitrogen and phosphorus absorption, thereby significantly increasing onion output. Furthermore, gibberellic acid (GA_3), a crucial plant growth controller, plays a key role in the onion's development cycle. When used skilfully, GA_3 promotes stem elongation, internode expansion and cell division, contributing to the development of larger bulbs. Humic acid, an organic substance, also plays a major part by enhancing soil structure, nutrient assimilation, root growth and water retention, all of which collectively lead to improved onion growth and better bulb quality.

The strategic and skilful use of these natural components is vital for maximizing onion farming results. By using bio-fertilizers and growth controllers, not only can quality be enhanced, but sustainable agricultural practices can also be promoted by reducing reliance on synthetic inputs. By harnessing the potential of these natural growth boosters, farmers can guarantee the sustainable and productive cultivation of onions, satisfying the growing demand for this versatile herb in various cooking customs.

MATERIALS AND METHODS

The research was conducted at Jawaharlal Nehru Krishi Vishwa Vidyalaya, JNKVV Jabalpur during Rabi season of 2022-23. The focus of the study was on the Bhima Shakti variety of onion. To conduct the study, a Randomized Complete Block Design was used with three replications and 12 treatments. The onions were grown in plots measuring 3x2 meters with 1-meter gaps between replications. The distance between the onions was 10 cm, while the distance between the rows was 15 cm. The objective of the study was to assess the effectiveness of a mixture of Azotobacter and Azospirillum, combined with 5 kg/ha of Farmyard Manure (FYM) during transplantation, in enhancing nitrogen fixation and nutrient availability. Furthermore, 2 kg/ha of humic acid and 100 ppm of Gibberellic acid (GA_3) were applied to the leaves 60 days after transplantation to improve nutrient absorption, promote growth and increase yield. The impact of these interventions was evaluated 90 days after transplantation.

T1 received RDF (NPKS:100:50:50:40 Kg/ha), T2 received RDF + Azotobacter (5kg/ha), T3 received RDF + Azospirillum (5kg/ha), T4 received RDF + GA_3 (100 ppm), T5 received RDF + Humic acid (2kg/ha), T6 received RDF + Azotobacter (5kg/ha) + Azospirillum (5kg/ha), T7 received RDF + GA_3 (100 ppm) + Azotobacter (5kg/ha), T8 received RDF + GA_3 (100 ppm) + Azospirillum (5kg/ha), T9 received RDF + GA_3 (100 ppm) + Humic acid (2kg/ha), T10 received RDF + GA_3 (100 ppm) + Azotobacter (5kg/ha) + Azospirillum (5kg/ha) + Humic acid (2kg/ha), T11 received RDF + Humic acid +

Azotobacter (5kg/ha) + Azospirillum (5kg/ha), and T12 served as the control group (Figure 1).

Table 1: **Overview of Treatments**

Treatments No.	Treatment Details
T_1	RDF (NPKS:100:50:50:40 Kg/ha)
T_2	RDF + Azotobacter (5kg/ha)
T_3	RDF + Azospirillum (5kg/ha)
T_4	RDF + GA_3 (100 ppm)
T_5	RDF + Humic acid (2kg/ha)
T_6	RDF + Azotobacter + Azospirillum
T_7	RDF + GA_3 + Azotobacter
T_8	RDF + GA_3 + Azospirillum
T_9	RDF + GA_3 + Humic acid
T_{10}	RDF + GA_3 + Azotobacter + Azospirillum + Humic acid
T_{11}	RDF + Humic acid + Azotobacter + Azospirillum
T_{12}	Control

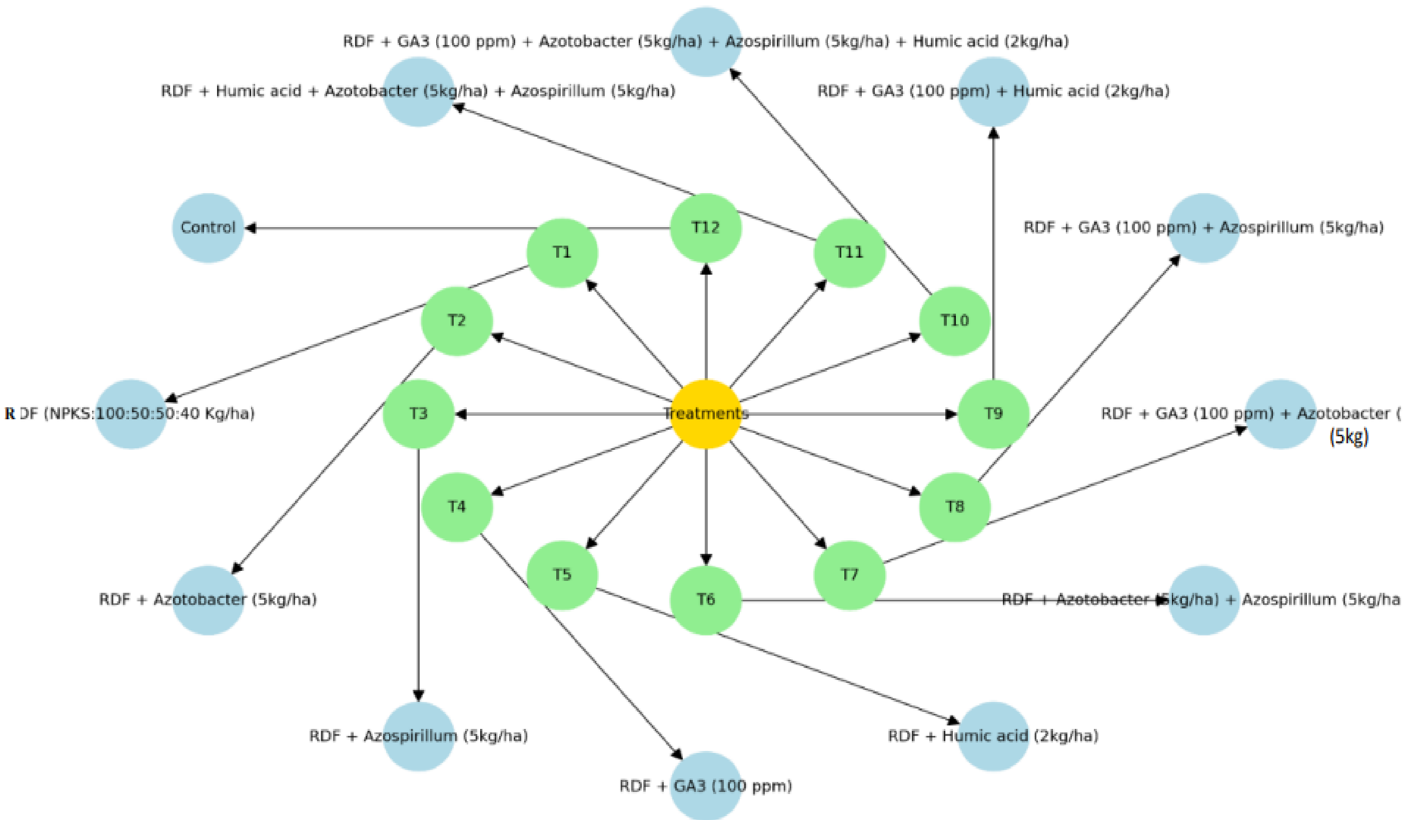

Figure 1: Treatments details

The observations for the following parameters were recorded using standard methodology:

Leaf Area per plant (cm^2)

Leaf area measure by leaf area meter.

Leaf Area Index (LAI)

LAI was measured with mainly based on leaf area divided by ground area occupied by plant.

Leaf area index is the ratio of leaf area to ground area. (5kg)

$$LAI = \frac{\text{Leaf Area}}{\text{Ground Area}}$$

TSS estimation

TSS or Total Soluble Solids, is quantified using a refractometer. This device is employed to measure the concentration of dissolved solids, often in the form of sugars, in a liquid sample, providing a valuable metric for assessing the sweetness and quality of substances like fruit juices and other beverages.

Pyruvic acid estimation

To determine the concentration of pyruvic acid, we mixed 0.5 ml of onion extract with 1.5 ml of 5% TCA and 18 ml of distilled water. After that, we combined 1ml of this mixture with 1ml each of 2,4-DNPH and distilled water, and let it incubate at 37ºC for 10 minutes. Then, we added 5 ml of 0.6 N NaOH and measured the absorbance at 420 nanometres using a Spectrophotometer to establish a standard curve. The concentration of pyruvic acid is a crucial indicator of onion pungency, determined by the breakdown of taste precursors. This helps in evaluating the flavour and sensory characteristics of the onions being studied.

Pungency range

The onions are divided into three categories based on their pungency: low, which is (0–3 µmol pyruvic acid/g); medium, which is (3–7 µmol pyruvic acid/g); and high, which is (greater than 7 µmol pyruvic acid/g). According to Bhima Shakti data, it can be categorized as medium/sweet pungent (3-7 µmol) (Dhumal et al., 2007) [5].

RESULTS AND DISCUSSION

After 60 days of planting, treatment T10, which consisted of a comprehensive combination of Recommended Dose of Fertilizer (RDF), Gibberellic Acid (GA_3), Azotobacter, Azospirillum and Humic acid, resulted in the tallest onion plants measuring 50.31 cm. This height was closely matched by T11 (49.68 cm), which used a similar mixture excluding GA_3. In contrast, the control group T12 had the shortest height at 41.92 cm. By 90 days after planting, T10 continued to lead with a height of 62.23 cm, closely followed by T11 at 61.47 cm, while T12 registered a height of 51.88 cm. The significant growth response to the combination treatments of T10 indicates its effectiveness in promoting cell elongation and root expansion, which is consistent with the findings of Sajid et al. (2012) and Mohammed and Ibraheem (2020) [13,11].

In terms of foliar characteristics, T10 exhibited superior leaf count, length, area and diameter. At 60 days after planting, T10 had the highest leaf count (7.15) and length (44.91 cm), followed closely by T11 with 6.95 leaves and a considerable leaf area. By 90 days after planting, T10 maintained its lead with a leaf count of 10.18 and a length of 54.08 cm, which were nearly equivalent to T11's 9.45 leaves and significant leaf area of 596.03 cm^2. On the other hand, T12 consistently showed the lowest values in all these attributes. When it comes to leaf area, T10 had the largest measurements at both 60 days after planting (320.69 cm^2) and 90 days after planting (624.65 cm^2), while T12 consistently recorded the smallest leaf area (241.15 cm^2 at 60 days after planting and 360.75 cm^2 at 90 days after planting). These trends highlight the positive influence of the treatment components on foliar development, which aligns with the findings of Singh et al. (2002), Gowda et al. (2007) and Kumar et al. (2017) [15,7,10].

The Leaf Area Index (LAI) showed similar patterns, with T10 demonstrating the highest values at both 60 and 90 days after transplanting (DAT), recording 2.13 and 4.16 respectively. T11 closely paralleled these figures with an LAI of 2.11 and 3.97, while T12 consistently had the lowest values (1.60 and 2.40). In terms of leaf diameter, T10 led with the largest measurements at 60 DAT (6.73 mm) and 90 DAT (8.14 mm), similar to T11's figures, while T12 consistently had the smallest diameters (Table 2 and Figure 2). These results emphasize the significant impact of the treatment components on plant growth and their importance in assessing LAI, which supports the observations of Calvo et al. (2014) and Singh et al. (2002) [2,15].

Table 2: Response of biofertilizer, GA_3 and humic acid on growth parameters

S. No.	Treatments	Plant height (cm)		Number of leaves/plant		Leaf length (cm)		Leaf area per plant (cm^2)		Leaf area index		Diameter of leaf (mm)	
		60 DAT	90 DAT	60 DAT	90 DAT	60 DAT	90 DAT	60 DAT	90 DAT	60 DAT	90 DAT	60 DAT	90 DAT
T1	RDF (NPKS: 100:50:50:40 Kg/ha)	46.91	56.68	5.85	7.09	41.61	48.66	261.98	445.14	1.74	2.96	5.49	6.99
T2	RDF + Azotobacter (5kg/ha)	48.90	58.11	6.09	7.87	42.63	49.74	310.37	501.26	2.06	3.34	5.98	7.09
T3	RDF + Azospirillum (5kg/ha)	48.15	57.88	5.99	7.80	42.9	49.85	290.67	450.40	1.93	2.99	5.88	7.15
T4	RDF + GA_3 (100 ppm)	47.09	59.25	6.21	8.52	41.57	50.59	272.99	505.83	1.81	3.37	5.85	7.20
T5	RDF + Humic acid (2kg/ha)	47.00	59.39	6.22	8.60	41.69	50.73	267.79	516.74	1.78	3.44	5.77	7.12
T6	RDF + Azotobacter + Azospirillum	49.09	59.84	6.23	8.80	43.74	51.59	317.14	533.16	2.09	3.55	6.20	7.56
T7	RDF + GA_3 + Azotobacter	48.11	60.15	6.74	8.87	42.67	52.65	285.77	591.05	1.90	3.93	6.18	7.46
T8	RDF + GA_3 + Azospirillum	48.13	60.13	6.42	8.88	42.50	52.58	290.22	546.06	1.93	3.64	5.96	7.75
T9	RDF + GA_3 + Humic acid	47.88	60.01	6.78	9.19	41.29	52.54	279.08	542.32	1.85	3.61	5.91	7.20
T10	RDF + GA_3 + Azotobacter + Azospirillum + Humic acid	50.31	62.23	7.15	10.18	44.91	54.08	320.69	624.65	2.13	4.16	6.73	8.14
T11	RDF + Humic acid + Azotobacter + Azospirillum	49.68	61.47	6.95	9.45	44.15	53.52	314.74	596.03	2.11	3.97	6.23	7.89
T12	Control	41.92	51.88	5.93	6.22	39.45	45.25	241.15	360.75	1.60	2.40	4.59	5.86
	Sem±		0.33		0.34		0.27		16.09		0.10		0.31
	CD at 5%		0.99		0.99		0.81		47.18		0.31		0.92

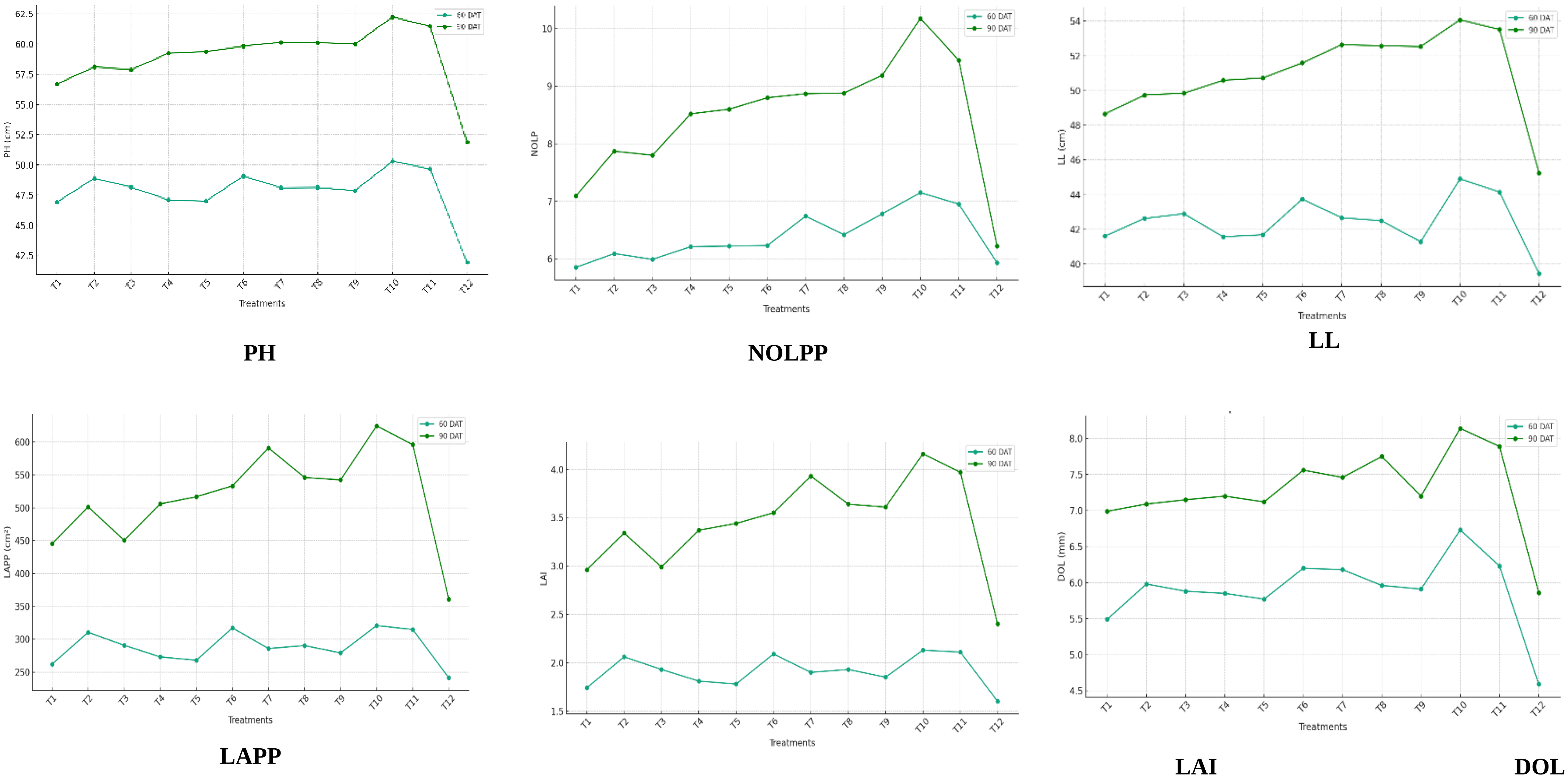

Figure 2: Comparison of PH (plant height), NOLPP (number of leaves per plant), LL (leaf length), LAPP (leaf area per plant), LAI (leaf area index), DOL (diameter of leaf) at 60 and 90 DAT (Days after Transplanting)

Quality parameters

The impact of different treatments, namely Azotobacter, Azospirillum, GA_3 and humic acid, on the Total Soluble Solids (TSS) content in onions is emphasized by this research. The results showed that Treatment T10, which consisted of RDF, GA_3, Azotobacter, Azospirillum and Humic acid, recorded the highest TSS value at 12.37 °Brix. Treatment T11, which included RDF, Humic acid, Azotobacter and Azospirillum, closely followed with a TSS value of 12.08 °Brix. Conversely, Treatment T12 (Control) had the lowest TSS value at 9.00 °Brix. These treatments significantly enhanced photosynthesis, optimized nutrient utilization and stimulated sugar synthesis, leading to an increase in the TSS content of Treatment T10. This finding supports previous research (Dilpreet et al., 2016 and Gupta et al., 2023) [4,8], highlighting the effectiveness of these treatments in increasing TSS levels in onions.

Furthermore, the study provides compelling evidence of the significant effect of these treatments on the pyruvic acid content in onion plants. Treatment T10 once again had the highest pyruvic acid content at 5.28 µmol/gm, followed closely by Treatments T11 and T9, which recorded 5.25 µmol/g and 5.24 µmol/g respectively and were statistically equivalent. In contrast, Treatment T12 (Control) had the lowest pyruvic acid content at 3.6 µmol/g. The inclusion of sulphur and humic acid played a crucial role in enhancing pyruvic acid metabolism, leading to increased energy production and improved cellular metabolic efficiency. This study aligns with previous findings by Denre et al. (2014) [3] and highlights potential opportunities for enhancing the nutritional characteristics of onions. Based on the data, Bhima Shakti onions are classified as having medium/sweet pungency, with a pyruvic acid content ranging from 3-7 µmol/g (Table 3 and Figure 3).

Table 3: Response of biofertilizer, GA_3 and humic acid on quality parameters

S. No.	Treatment	TSS (°Brix)	Pyruvic acid (µmol/g)	Pungency
T1	RDF (NPKS: 100:50:50:40 Kg/ha)	10.04	4.12	Medium pungent
T2	RDF + Azotobacter (5kg/ha)	10.76	4.09	medium pungent
T3	RDF + Azospirillum (5kg/ha)	10.34	4.10	medium pungent
T4	RDF + GA_3 (100 ppm)	10.10	4.22	medium pungent
T5	RDF + Humic acid (2kg/ha)	10.17	5.12	medium pungent
T6	RDF + Azotobacter + Azospirillum	11.23	4.23	medium pungent
T7	RDF + GA_3 + Azotobacter	11.03	4.13	medium pungent
T8	RDF + GA_3 + Azospirillum	11.00	4.20	medium pungent
T9	RDF + GA_3 + Humic acid	10.64	5.24	medium pungent

T10	RDF + GA_3 + Azotobacter + Azospirillum + Humic acid	12.37	5.28	medium pungent
T11	RDF + Humic acid + Azotobacter + Azospirillum	12.08	5.25	medium pungent
T12	Control	9.00	3.06	medium pungent
	Sem±	0.32	0.10	
	CD at 5%	0.96	0.30	

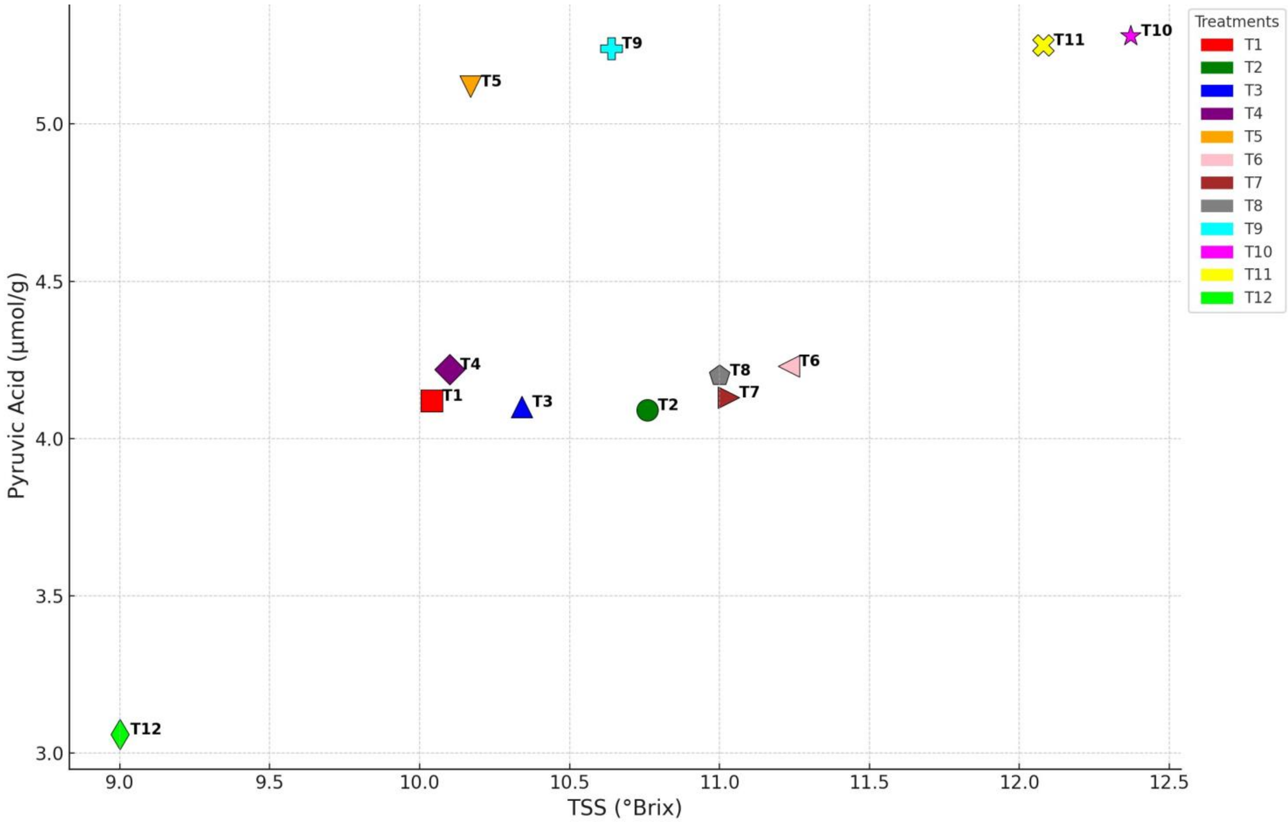

Figure 3: Custom visualization of TSS and pyruvic acid with annotations

CONCLUSION

To achieve exceptional growth and unmatched quality in cultivating the Bhima Shakti onion variety by judicial application of different treatment approaches. Particularly the combined use of Azotobacter (5 kg), Azospirillum (5 kg), GA_3 (100 ppm) and humic acid (2 kg) representing the treatment T10 demonstrated an impressive growth include reaching an ideal plant height of 62.23 cm, maximizing the number of leaves per plant to 10.18, maintaining an average leaf length of 54.08 cm, achieving a considerable leaf area of 624.65 cm^2 per plant, promoting a strong Leaf Area Index (LAI) of 4.16 and securing a sufficient leaf diameter of 8.14 mm. At the same time, it is essential to address quality indicators such as Total Soluble Solids (TSS) at 12.37 °Brix and pyruvic acid concentrations at 5.28 µmol/g to ensure a harvest of exceptional quality.

ACKNOWEDGEMENTS

The authors express their sincere gratitude to the Department of Horticulture at the College of Agriculture, Jawaharlal Nehru Krishi Vishwa Vidyalaya (JNKVV), Jabalpur, Madhya Pradesh, India. I also express my gratitude to Dr. Manoranjan Biswal for statistical analysis and visualization.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Aswani G, Paliwal R and Sarolia DK. Effect of nitrogen and bio-fertilizer on yield and quality of rabi onion (*Allium cepa* L.) CV. Puna Red. Agric. Sci. Digest. 2005;25 (2):124-126.
2. Calvo P, Nelson L and Kloepper JW. Agricultural uses of plant biostimulants. Plant and Soil. 2014;383:3-41.
3. Denre M, Ghanti S and Sarkar K. Effect of humic acid application on accumulation of mineral nutrition and pungency in garlic (*Allium sativum* L.). International Journal for Biotechnology and Molecular Biology Research. 2014; 5(2):7-12.
4. Dilpreet, Talwar, Singh, Kulbir, Sardana, Varinder Singh and Jagdis. Growth, yield and quality of onion as influenced by integrated nutrient management. International Journal of Agriculture Sciences. 2016;8(51):2295- 2298.
5. Dhumal K, Datir S and Pandey R. Assessment of bulb pungency level in different Indian cultivars of onion (*Allium cepa* L.). Food Chem. 2007;100(4):1328–1330.
6. Fawy HA, Ibrahim SM, Abo EL-Ela H K and Abd EL- Hameed NM. The integration effect between mineral and biofertilization on wheat production in soils high in iron at the New Valley, Egypt. Egypt. J. Soil Sci. 2016;56(1):53-68.
7. Gowda CM, Vijaykumar M and Gowda Mallikarjuna AP. Influence of integrated nutrient management on growth, yield and quality of garlic (*Allium sativum* L.) cv. G-282. Crop Research. 2007;33(1-3):144-147.
8. Gupta P, Trivedi J, Sharma PK, Devi S and Siwna Y. Effect of different bio-fertilizers and organic substances on quality parameters of rabi onion (*Allium cepa* L.). The Pharma Innovation Journal 2023;12(4):209-212.
9. Kumar A, Ram RB, Maji S, Kishor S, Yadav R, Govind and Meena KR. Effect of organic manures, biofertilizers and micronutrients on growth, yield and quality of onion (*Allium cepa* L.). International Journal of Agricultural Sciences 2017;13(2):236-241.

10. Kumar A, Meena ML, Shivran BC, Pal H and Meena BL. Impact of Bio-fertilizer on growth, yield and quality of onion (*Allium cepa* L.) CV. Pusared. Plant Archives. 2019;19(1):772-776.
11. Mohammed AS and Ibraheem FFR. Response of onion plant to the humic acid application and spraying with liquid fertilizer on growth and yield of green onion. Plant Archives. 2020;20(1):1141-1144.
12. Roldan E, Sanchez-Moreno C, de Ancos B and Cano MP. Characterization of onion (*Allium cepa* L.) by-products as food ingredients with antioxidant and antibrowning properties. Food Chem. 2008;108:907-916.
13. Sajid M, Rab A, Shah ST, Jan I, Haq I, Haleema B, Zamin M, Alam R and Zada H. Humic acids affect the bulb production of onion cultivars. African Journal of Microbiology Research 2012;6(28):5769-5776.
14. Sekara A, Pokluda R, Del Vacchio L, Somma S and Caruso G. Interactions among genotype, environment and agronomic practices on production and quality of storage onion (*Allium cepa* L.)—A review. Hort. Sci. (Prague). 2017; 44:201-212.
15. Singh P, Tiwari N and Katiyar PK. Pre transplant seedling treatment with growth regulator and their effect on the growth and bulb production of onion (*Allium cepa* L.). Progressive Agriculture 2002;2(2):181-182.
16. Wu SC, Cao ZH, Li ZG and Chung KC. Effect of biofertilizer containing N2 -fixen, P and K solubilizers and A. M. fungi on maize growth: A greenhouse trail. Geoderma. 2005;125:155-166.