



RESEARCH ARTICLE

Slow-release fertilizers improve micronutrient uptake and boost morphological, physiological and nutritional traits in tomato

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Received: 10 May 2025; Accepted: 02 July 2025; Available online: Version 1.0: 14 September 2025; Version 2.0: 06 October 2025

Cite this article: Abreeta K, Elayarajan M, Janaki P, Suganya S, Sankari A, Parameswari E, Behera SK. Slow-release fertilizers improve micronutrient uptake and boost morphological, physiological and nutritional traits in tomato. Plant Science Today. 2025;12(sp1):01–08. <https://doi.org/10.14719/pst.9402>

Abstract

Slow-Release Fertilizers (SRFs) provide substantial environmental advantages over conventional fertilizers by mitigating nutrient losses via leaching, volatilization and surface runoff. By synchronizing nutrient release with plant uptake, SRFs enhance nutrient use efficiency while minimizing the risk of soil and water contamination. The development of SRFs plays a pivotal role in reducing the environmental footprint and enhancing the yield and quality of crops while minimizing nutrient losses. Keeping all this points in mind an attempt was made to develop a SRF using four essential micronutrients- zinc (Zn), iron (Fe), copper (Cu) and manganese (Mn) based on crop's nutrient requirement which was coated with different polymers comprising a total of 12 treatments. A field trial was conducted to evaluate the results during the *Rabi* season (November to February) in Tamil Nadu. The results demonstrated that treatment T₈ (MNF coated with biochar + humic acid @ 28 kg ha⁻¹) followed by T₇ (MNF coated with biochar + humic acid @14 kg ha⁻¹) significantly enhance the growth, chlorophyll content and quality parameters of tomato compared to uncoated (T₃ & T₄) and absolute control plot (T₁). This study concludes that SRFs can provide a solution to effective nutrient release. Further, it draws attention towards use of biochar and humic acid in attaining marked improvements highlighting their potential to meet crop nutritional demands efficiently over time.

Keywords: biochar; humic acid; micronutrients; nutrient use efficiency

Introduction

By the end of 2050, the world's population is predicted to have grown to 9 billion people at a rate of 1.05 % (1). As the world's population grows, there will be a 60 % rise in food consumption above current levels (2,3). The loss of fertile topsoil due to intensive cropping, inadequate organic matter addition, heavy metal contamination and insufficient micronutrient supplementation is causing serious problems. Additionally, a lack of micronutrients is increasingly making it difficult to get the best yield. Since they regulate most enzymatic reactions, these micronutrients are vital. Their absence in plants led to a notable decrease in yield characteristics (4). Plants use micronutrients for a variety of purposes, including as cofactors of antioxidant enzymes and as structural elements in osmolites under stress, which are essential for plant growth and development (5,6). Furthermore, it is commonly known that a reduction in plant performance and yield due to micronutrient deficiencies negatively impact sustainable agriculture (7). The formation of agricultural plants depends on microelements including manganese, iron, zinc and copper, which are needed in trace

levels (8). For plants to meet the world's food needs, fertilisers are a vital supply of nutrients.

Globally, the tomato (*Solanum lycopersicum*) is the third most produced crop. In India, tomato is grown throughout the country. Tomatoes are frequently consumed, either as raw fruit or in processed form, because of their high mineral, vitamin and anti-cancer properties (9). Balanced nutrition through organic manures is advocated; however, they may only support low yield levels. For harnessing the higher yield potential, supplementation of micronutrients is essential. Amongst the vegetables, tomato is very responsive to the application of micronutrients. The micronutrients improve the chemical composition of fruits and general condition of plants and are known to act as catalyst in promoting organic reactions taking place in plants (10). Applications of micro-nutrients i.e., zinc and boron have been reported in increasing growth and seed yield in tomato. When farmers utilise micronutrient fertilisers to increase output, tomatoes (*Solanum lycopersicum*) are the most economical vegetable (11).

SRFs application is advised to get around problems with conventional fertiliser such as fertility loss from indiscriminate use, groundwater pollution and environmental stress (12). SRFs demonstrated a more secure, cost-effective and effective method of supplying fertilisers to satisfy crop needs. SRFs are defined as a fertilizer coated with an organic/inorganic compound aimed at sustaining the nutrient release and increase the duration for nutrient availability to the crops significantly due to presence of polymeric coating material (13). SRFs are made to satisfy crops' nutrient needs according to their growth cycle (10). Fertiliser is used as the core material in SRF manufacturing and coating materials such as resin or polymers are used to shield the core and release the core nutrients in a regulated way (14,15). Considering the urgent need for food security and sustainable development, the importance of introducing effectively formulated fertilizers is nowadays very noticeable and severe. Therefore, many efforts have been focused on offering environmentally benign coatings to produce the SRFs (16-18).

The present work aims to evaluate different SRF formulations developed using combination of different organo-monomers (19) used for coating the MNF granules at two different levels (14 and 28 kg ha⁻¹) and validate the best treatment under field conditions. Therefore, keeping the above points, this study was conducted with an aim to improve the morphological, physiological and nutritional parameters of tomato (*S. lycopersicum* L.) crop using the different organo-monomeric coated SRFs at two different levels under field condition.

Materials and Methods

Study area and experimental design

The experiment was conducted in Ettimadai town of Coimbatore district. The experimental sites were located at an elevation of 313 m above mean sea level, with the main site having geographical coordinates of latitude 10.8900° N and longitude 76.9088° E. The study aimed to assess and develop SRF products under various treatments to manage and increase the micronutrient uptake, content along with growth, yield and quality parameters of tomato crop. The experimental layout followed a Randomized Block Design (RBD), with three replications assigned per treatment to account for environmental variability. Each treatment was applied to plots of equal dimensions, measuring 3 m by 5 m, with buffer zones of 25 cm between plots to minimize cross-contamination.

Soil characteristics and fertilizer application

The initial surface soil analysis of the experimental field revealed the following characteristics: a clayey loam texture with 22.08 % coarse sand, 24.41 % fine sand, 20.6 % silt and 32.5 % clay estimated using international pipette method. The bulk density of 1.25 Mg m⁻³, particle density of 2.65 Mg m⁻³ and porosity of 50 % was recorded using cylinder method. The pH of the soil was recorded as 7.62 using 1:2.5 soil: water suspension potentiometry, with an Electrical Conductivity (EC) of 0.16 dS m⁻¹ using 1:2.5 soil: water suspension conductimetry and a Cation Exchange Capacity (CEC) of 32.5 C mol (p⁺) kg⁻¹. The soil had a free CaCO₃ content of 2.75 % which was estimated using rapid titration method, organic carbon content of 0.75 % estimated through wet digestion method,

available KMnO₄-N at 210 kg ha⁻¹ estimated by alkaline permanganate method Olsen-P at 16 kg ha⁻¹ estimated using spectrophotometer and NH₄OAc-K at 392 kg ha⁻¹ estimated through flame photometer. The DTPA-extractable micronutrients were measured as follows: zinc (3.93 mg kg⁻¹), copper (2.18 mg kg⁻¹), iron (3.44 mg kg⁻¹) and manganese (1.08 mg kg⁻¹) which were estimated using 0.005 M DTPA extractant as outlined by Lindsay and Norvell method.

Preparation of SRF by rotary drum method

The micronutrient formulation was made with respect to the Tamil Nadu Agricultural University (TNAU) micronutrient recommendation given in Crop Production Guide (CPG) and according to that the respective fertilizers such as zinc sulphate, iron sulphate, copper sulphate and manganese sulphate were mixed in a specific ratio. Then with addition of potato dextrose as a binder the mixture was further rotated at 360 rpm for 30 min which gave it a spherical granular shape. Those granules were further coated with biochar (coconut husk pyrolyzed under a temperature of 600 °C) in 2:1 ratio and after drying it was further double coated with Humic Acid (HA) (6 % w/v), Cassava Starch (CS) (6 % w/v).

Likewise, the micronutrient granules were also coated with bentonite using 2:1 ratio and further double coated with HA and with CS at 6 % w/v. All the SRFs were tested at two levels (14 and 28 kg ha⁻¹) of fertilizer application along with uncoated and absolute control treatments. The tomato variety used was PKM-1 with a row spacing of 45 cm by 30 cm to ensure optimal plant stand. Standard agronomic practices, including irrigation, weed management and pest control, were uniformly implemented across all plots. Irrigation was applied as required and all cultural operations were performed to promote ideal crop growth.

The treatments comprised a total of 12 numbers of details of which are as follows in Table 1.

Plant height measurement

Plant height (cm) was monitored regularly throughout the crop growth period to evaluate the effect of each treatment. Five tomato plants were randomly selected from each plot, excluding border rows. The height of each plant was measured from the soil surface to the tip of the tallest leaf. These measurements were recorded at different growth stages and the average plant height for each plot was calculated.

Table 1. Treatment details

Treatment	Details
T ₁	Absolute control (No MN)
T ₂	MN applied as per TNAU recommendation
T ₃	Uncoated MNCF granules @ 14 kg ha ⁻¹
T ₄	Uncoated MNCF granules @ 28 kg ha ⁻¹
T ₅	Biochar+CS @ 14 kg ha ⁻¹
T ₆	Biochar+CS @ 28 kg ha ⁻¹
T ₇	Biochar+HA @ 14 kg ha ⁻¹
T ₈	Biochar+HA @ 28 kg ha ⁻¹
T ₉	Bentonite+CS @14 kg ha ⁻¹
T ₁₀	Bentonite+CS @ 28 kg ha ⁻¹
T ₁₁	Bentonite+ HA @14 kg ha ⁻¹
T ₁₂	Bentonite+ HA @14 kg ha ⁻¹

Root length

Root length (cm) was monitored regularly throughout the crop growth period to evaluate the effect of each treatment. Five tomato plants were randomly selected from each plot, excluding border rows. The length of the primary root from each plant was measured from the point of attachment of the crop to the tip. These measurements were recorded at different growth stages and the average root length for each plot was calculated.

SPAD

The SPAD chlorophyll meter was used to evaluate chlorophyll concentration, indicating the photosynthetic potential and overall plant health. Chlorophyll content was measured in the physiologically active leaves of the five labelled plants at various growth stages in each plot. The SPAD readings were averaged for each plot and these values provided insights into how nutrient availability from different fertilizer treatments influenced plant health (20).

Enzymatic activity

Reaction mixture of pyrogallol and hydrogen peroxide without the leaf extract constituted the blank. 500 mg of fresh leaves was homogenized with 0.1 M phosphate buffer and centrifuged at 5000 rpm for 15 min in cool condition. 1 mL of supernatant, 3 mL of 0.05 M pyrogallol and 0.5 mL of 30 % H₂O₂ were taken in a test tube. The change in absorbance was measured at 430 nm for every 30 sec up to 150 sec and the enzyme activity was calculated and expressed as unit g FW⁻¹ of sample.

Data analysis

The collected data on plant growth parameters, yield and quality attributes were subjected to analysis of variance (ANOVA) to evaluate the statistical significance of the differences between treatments. Mean values were compared using the Least Significant Difference (LSD) test at a 5 % significance level. All statistical analyses were conducted using GRAPES (General Rshiny Based Analysis Platform Empowered by Statistics) software version 1.0.0 and graphical representations were generated to illustrate the effects of the various treatments.

Results

The findings show that tomato plant height rose gradually across all the growth stages, starting from vegetative, flowering, reproductive and harvest stages. Table 2 shows the noteworthy differences between various treatments with T₈ (biochar + HA coated @ 28 kg ha⁻¹) emerging as the best treatment followed by T₇ (biochar + HA coated @ 14 kg ha⁻¹) while T₁ (control without any micronutrients) consistently recorded the lowest values among all treatments. T₈ recorded a plant height of 54.5 cm during the vegetative stage followed by T₇ 49.46 cm and T₁ 24.93 cm. T₈ continued with this increasing trend and measured 71.00 cm, 89.00 cm and 108.33 cm during the flowering, reproductive and harvest stages respectively. This was followed by T₇ with showed the measurements as 67.33 cm, 84.66 cm and 95.66 cm while T₁ showed 37.13 cm, 51.66 cm and 65.90 cm respectively for flowering, reproductive and harvest stages of the crop. The SE(D) and C.D. values (5 %) showed that the difference between the treatments were significant statistically which has been indicated by different letters along a column.

Consequently, root length also showed a similar trend and T₈ emerged as the best treatment. At vegetative stage it was observed that maximum root length of 10.20 cm belonged to T₈ followed by T₇ (9.60 cm) and least was for T₁ (6.42 cm). Similarly, for flowering, reproductive and harvest stages it was found to follow the same trend with T₈ having values 17.32 cm, 20.20 cm and 25.49 cm corresponding for flowering, reproductive and harvest stages respectively. T₇ had values as 16.78 cm, 19.30 cm and 24.40 cm belonging to flowering, reproductive and harvest stages whereas for T₁ observed values were 8.10 cm, 9.80 cm and 15.20 cm belonging to flowering, reproductive and harvest stages. It was observed that T₈ emerged as the most effective treatment followed by T₇ and least with T₁ treatments T₉, T₁₀, T₂ and T₃ were found to be statistically insignificant (Table 2).

Comparably the chlorophyll content also gave us similar result with T₈ as the best treatment for all the stages of growth namely vegetative (65.5), flowering (68.8), reproductive (70.60) and harvest (71.40) as shown in Table 3. This was followed by T₇ with values starting from 63.19 (vegetative), 66.72 (flowering), 68.5 (reproductive) and 69.8 (harvest) stages. T₁ was found to have SPAD values with vegetative stage at 38.40, flowering stage (40.39), reproductive stage (44.59) and harvest stage at 46.39. These findings demonstrate well that the application of micronutrients is critical to plant's optimal functioning and contributing to photosynthetic activity as well. Use of coated fertilizers can give superior results as compared to uncoated fertilizers.

Consequently, enzymatic activity was also evaluated at successive stages for the crop and it was found that T₈ emerged superior for all the growth stages of the crop starting with vegetative (64.4), flowering (70.8), reproductive (77.2) and harvest (72.6) corresponding for peroxidase activity as indicated in Table 4. It was found that there is a gradual increase in the values starting from vegetative up to reproductive and further it declines at harvest stage. Treatment T₇ emerged as second best with values being recorded as 60.6,

Table 2. Effect of different treatments on plant height and root length as observed during various growth stages

	VS	FS	RS	HS	VS	FS	RS	HS
T ₁	24.93 ^j	37.13 ^j	51.66 ^j	65.90 ^l	6.42 ^j	8.10 ^j	9.80 ^j	15.20 ^j
T ₂	27.10 ⁱ	40.53 ⁱ	56.23 ⁱ	72.70 ⁱ	7.08 ⁱ	9.39 ⁱ	11.20 ⁱ	16.40 ⁱ
T ₃	28.03 ⁱ	41.96 ⁱ	57.20 ⁱ	73.83 ⁱ	7.25 ⁱ	9.64 ⁱ	11.66 ⁱ	16.39 ⁱ
T ₄	30.66 ^h	45.46 ^h	62.00 ^h	76.26 ^h	7.65 ^h	11.87 ^h	12.80 ^h	17.50 ^h
T ₅	33.23 ^g	49.16 ^g	64.80 ^g	78.53 ^g	7.91 ^g	12.49 ^g	14.20 ^g	18.60 ^g
T ₆	36.03 ^f	52.90 ^f	67.96 ^f	80.93 ^f	8.28 ^f	13.12 ^f	15.00 ^f	19.20 ^f
T ₇	49.46 ^b	67.33 ^b	84.66 ^b	95.66 ^b	9.60 ^b	16.78 ^b	19.30 ^b	24.40 ^b
T ₈	54.50 ^a	71.00 ^a	89.00 ^a	108.33 ^a	10.20 ^a	17.32 ^a	20.20 ^a	25.49 ^a
T ₉	38.56 ^e	55.50 ^e	70.43 ^e	84.53 ^e	8.56 ^e	11.32 ^e	15.80 ^e	20.40 ^e
T ₁₀	39.83 ^e	56.00 ^e	71.60 ^e	85.73 ^e	8.62 ^e	11.45 ^e	16.30 ^e	20.00 ^e
T ₁₁	43.16 ^d	59.93 ^d	75.56 ^d	89.06 ^d	8.90 ^d	16.00 ^d	17.20 ^d	21.80 ^d
T ₁₂	45.76 ^c	62.66 ^c	79.00 ^c	91.66 ^c	9.20 ^c	15.43 ^c	18.40 ^c	23.20 ^c
SE(d)	0.715	0.721	1.269	0.753	0.124	0.266	0.316	0.417
C.D.	1.483	1.495	2.632	1.562	0.257	0.551	0.655	0.866

VS - Vegetative stage; FS - Flowering stage; RS - Reproductive stage; HS Harvest stage.

Treatment means within column with different letters are significantly different

Table 3. Effect of different treatments on chlorophyll content as observed during various growth stages and fruit nutrient uptake at harvest stage

Treatments	SPAD			
	VS	FS	RS	HS
T ₁	38.40 ^l	40.39 ^l	44.59 ^l	46.39 ⁱ
T ₂	40.20 ^k	42.40 ^k	46.60 ^k	48.39 ^h
T ₃	42.80 ^j	44.60 ^j	49.60 ^j	50.59 ^g
T ₄	45.20 ⁱ	48.40 ⁱ	52.59 ⁱ	54.10 ^f
T ₅	47.79 ^h	50.40 ^h	54.30 ^h	55.60 ^f
T ₆	49.59 ^g	53.60 ^g	58.49 ^g	60.60 ^e
T ₇	63.19 ^b	66.72 ^b	68.50 ^b	69.80 ^a
T ₈	65.5 ^a	68.80 ^a	70.60 ^a	71.40 ^a
T ₉	52.60 ^f	55.40 ^f	60.80 ^f	62.40 ^d
T ₁₀	55.80 ^e	58.40 ^e	62.80 ^e	63.60 ^d
T ₁₁	57.40 ^d	60.40 ^d	64.29 ^d	65.59 ^c
T ₁₂	60.19 ^c	63.79 ^c	66.39 ^c	67.67 ^b
SE(D)	0.423	0.585	0.453	0.78
C.D.	0.876	1.213	0.94	1.618

VS - Vegetative stage; FS - Flowering stage; RS - Reproductive stage; HS Harvest stage.

Treatment means within column with different letters are significantly different

68.6, 75.0 and 70.7 correlating to vegetative, flowering, reproductive and harvest stages respectively. It was likely that T₁ was having the minimum values for all the stages namely vegetative (23.60), flowering (28.22), reproductive (44.20) and harvest (39.8).

Discussion

The results showed that biochar and HA can load nutrients and releasing them gradually over time, significantly influencing the morphological, physiological and quality parameters of tomato. In terms of plant height, root length, chlorophyll content and enzymatic activity treatment T₈ performed better than control and other fertilizers as indicated in Fig. 1a & 1b. These findings align with results and trials conducted with slow-release fertilizer on tomato crop which emphasizes how SRFs boosts the microbial diversity, improve nutrient holding capacity and soil structure and concurrently increase the nutrient availability which contribute to better crop performance (21). The gradual and continuous release of nutrients from the porous structure of the coated fertilizers is

Table 4. Effect of different treatments on peroxidase activity and polyphenol activity as observed during various growth stages

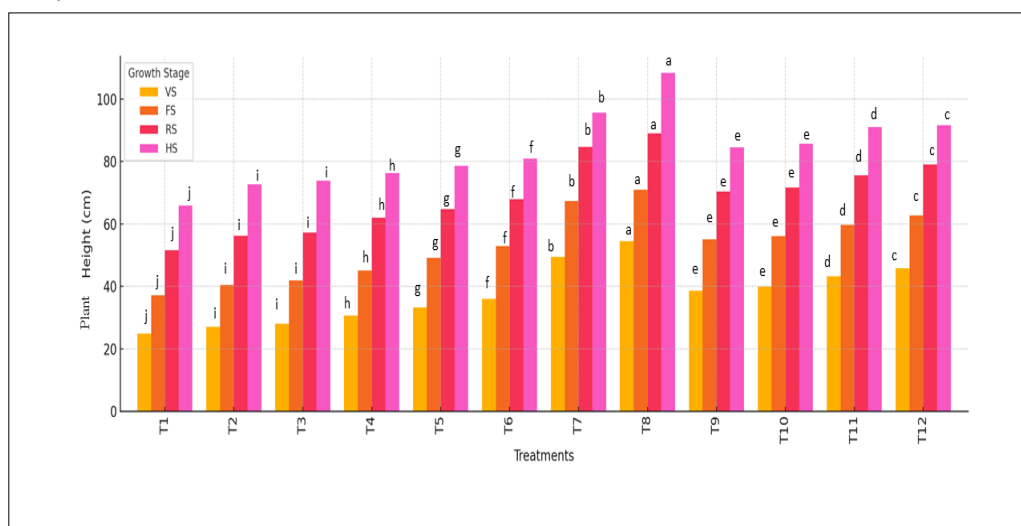
Treatments	Peroxidase activity			
	VS	FS	RS	HS
T ₁	23.60 ^l	28.22 ^k	44.20 ^l	39.80 ^k
T ₂	28.59 ^k	32.79 ^j	47.30 ^k	42.10 ^j
T ₃	32.80 ^j	36.61 ⁱ	52.80 ^j	44.69 ⁱ
T ₄	37.76 ⁱ	42.70 ^h	56.40 ⁱ	48.70 ^h
T ₅	40.00 ^h	46.30 ^g	60.19 ^h	52.20 ^g
T ₆	43.79 ^g	50.11 ^f	63.70 ^g	55.80 ^f
T ₇	60.60 ^b	68.60 ^b	75.00 ^b	70.70 ^b
T ₈	64.40 ^a	70.80 ^a	77.20 ^a	72.60 ^a
T ₉	47.80 ^f	53.20 ^e	66.30 ^f	58.40 ^e
T ₁₀	50.60 ^e	57.40 ^d	68.90 ^e	61.30 ^d
T ₁₁	53.40 ^d	65.50 ^c	71.20 ^d	67.50 ^c
T ₁₂	57.80 ^c	66.57 ^c	73.20 ^c	69.40 ^b
SE(D)	0.438	0.87	0.732	0.675
C.D.	0.909	1.804	1.519	1.4

VS - Vegetative stage; FS - Flowering stage; RS - Reproductive stage; HS Harvest stage.

Treatment means within column with different letters are significantly different

responsible for the noteworthy distinctions in terms of plant height, root length and chlorophyll content.

T₈ had the maximum plant height, root length, SPAD and enzyme activity for all stages as compared to control (T₁). These results are comparable to previous studies that combined application of micronutrients like zinc, boron and iron results in a synergistic effect, leading to a more pronounced increase in plant height compared to individual applications. This combination enhances various metabolic activities, promoting vigorous vegetative growth (22-24). On the other hand, lowest values were observed in T₁ (control) highlighting the essence of micronutrients for the enhanced growth and development. In terms of root growth and development it was found that T₈ consisting of biochar and HA coated fertilizer showed the highest value which can be attributed to the fact that HA can stimulate H⁺-ATPase activity in roots, promoting ion uptake and growing. Both biochar and HA are known to improve porosity, soil aeration and structure which promotes healthier nutrient uptake reducing the stress on roots and better organ development (23,24).

**Fig. 1a.** Plant height of tomato as recorded at various physiological stages.

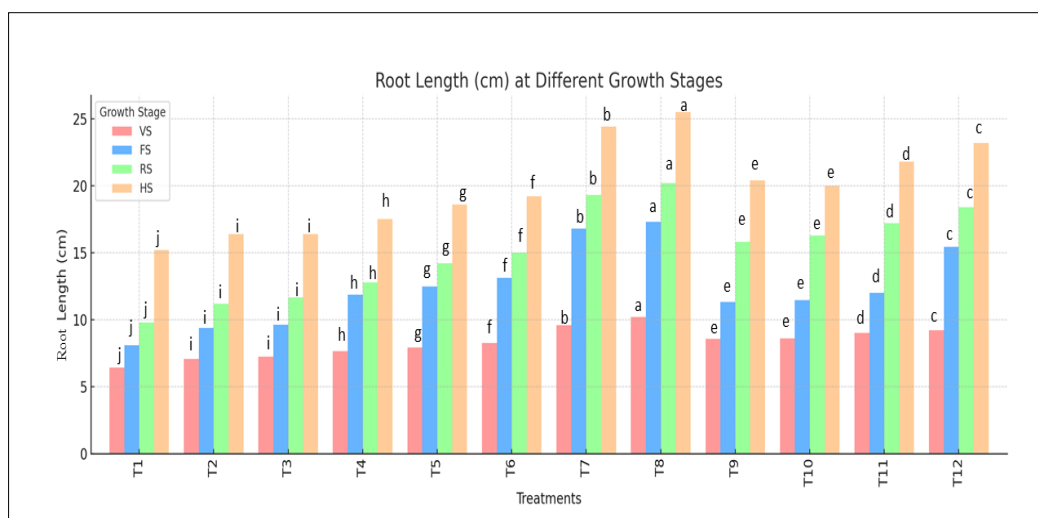


Fig. 1b. Root length of tomato as recorded at various physiological stages.

A comparison between T₇ and T₈ revealed that the higher fertilizer application rate in T₈ (28 kg ha⁻¹) led to superior growth compared to T₇ (14 kg ha⁻¹). However, T₇ performed better than the other treatments, likely due to the presence of biochar and HA coated fertilizer. T₁ had the least value which shows that though often neglected but micronutrient application is critical to crop growth and development.

T₈ also had the most significant SPAD values as shown in Fig. 2, which indicate chlorophyll concentration and photosynthetic efficiency. It has been demonstrated that biochar and HA coated micronutrients protect it from leaching or binding to other agents and improve their uptake by the crop. Chlorophyll requires micronutrients like iron, zinc, manganese as cofactors in biosynthetic pathways and

therefore its availability improves the SPAD values (25-27). The absence of these nutrients reduces the photosynthetic capability as demonstrated by the consistently low values throughout all growth stages for T₁ (control). Additionally, treatments containing other coated fertilizers also performed well than the control. Overall, the application of SRFs in combination with biochar induced increments in morphological and physiological parameters of tomato (28).

On the other hand, when we see antioxidant activity such as peroxidase, we observe that T₈ outperform the rest in all the growth stages of the crop followed by T₇ and the least performing treatment was T₁ (Fig. 3). This is because micronutrients play a significant role as enzyme cofactors. Iron is essential for heme group present in peroxidase and without

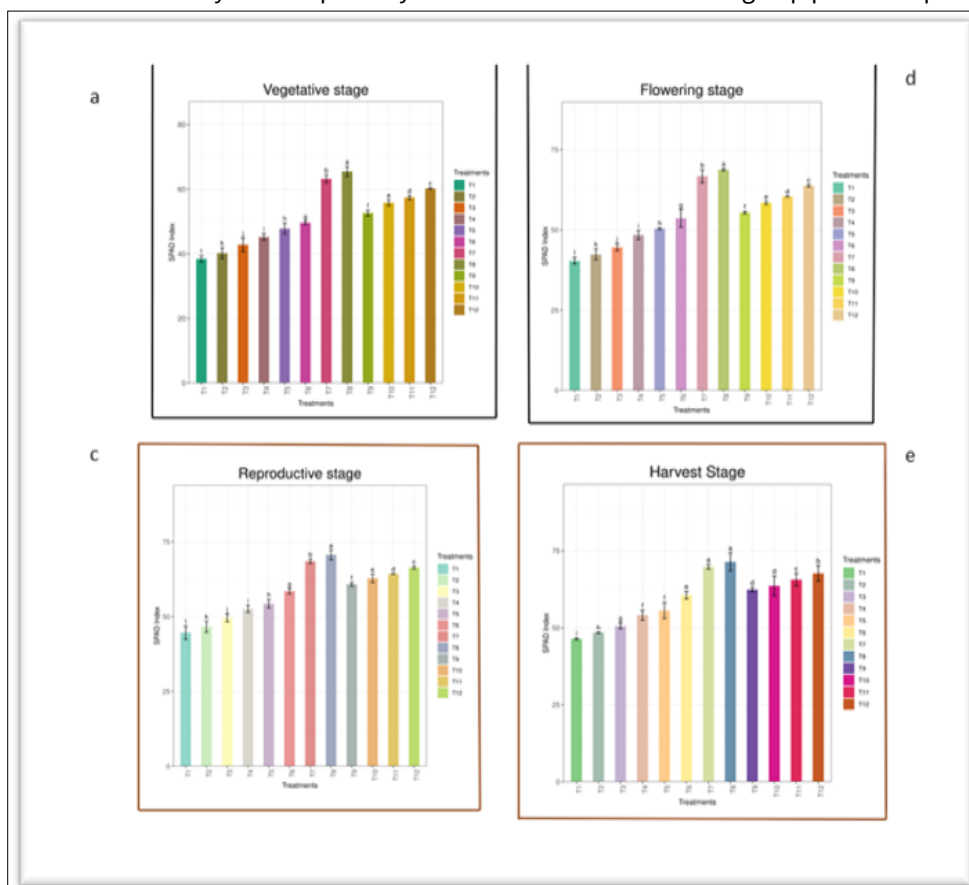


Fig. 2. Chlorophyll content (SPAD) as recorded at different physiological stages.

a. Vegetative stage; b. Flowering stage; c. Reproductive stage; d. Harvest stage

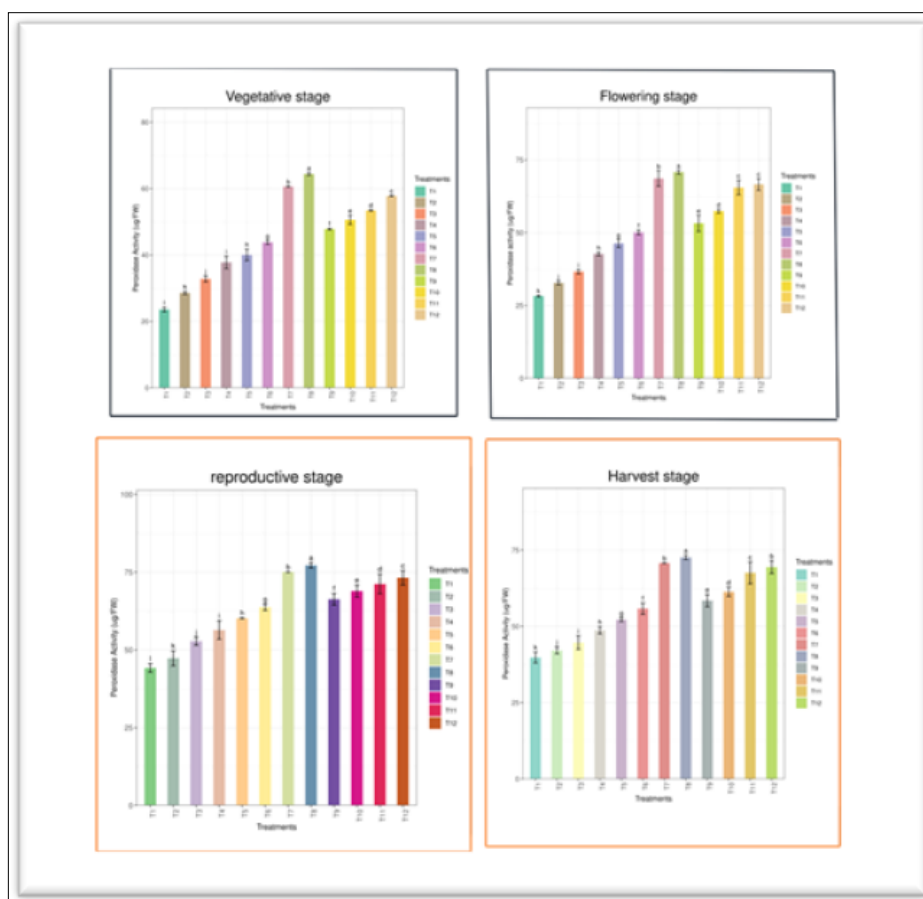


Fig. 3. Enzymatic activity as recorded at different physiological stages. a. Vegetative stage; b. Flowering stage; c. Reproductive stage; d. Harvest stage

it; enzyme's active site cannot bind with H_2O_2 and catalyse its breakdown. Likewise, copper stabilizes the enzyme structure and enables electron structure in enzymatic reactions. Similarly, zinc and manganese are also involved indirectly in holding the membrane integral and protein structures under oxidative stress. Therefore, deficiency of any micronutrient may lead to significant loss and reduction in peroxidase activity in the crop (25,29). Henceforth, it can be suggested that the application of micronutrients, whether through traditional fertilization or innovative approaches like slow/controlled systems delivery can significantly enhance peroxidase activity and overall antioxidant capacity in tomato plants.

Conclusion

The use of biochar and HA as coating materials for slow-release fertilizer development has demonstrated significant agronomic and nutritional benefits which was represented by treatment T_8 followed by T_7 . This study presents that the utilization of biomass derived biochar and HA coated SRFs are a novel approach for better utilization and uptake of micronutrients which often remains a subject of quite negligence. Additionally, their porous and functionalized surfaces enable gradual nutrient desorption, synchronizing nutrient release with plant uptake demand. The incorporation of biochar with micronutrients in the core and coating with HA showed positive impact in sustained release. This was evident from the uniform increments visible in morphological and physiological parameters such as plant height, root length, chlorophyll content and antioxidant activity throughout the growth stages of the crop. It is more likely that biochar seems to influence the

mechanical properties and hydrophobicity of the formulation and micronutrient release behaviour through the coated fertilizer. Therefore, biochar and HA are effective, sustainable and synergistic carriers for the controlled release of nutrients in modern agricultural systems.

Acknowledgements

Thanking the Department of Soil Science and Agricultural Chemistry, Tamil Nadu Agricultural University for providing financial support in carrying out the field trial and for all the time-to-time assistance required during the research work.

Authors' contributions

AK was responsible for writing and formulating the original draft. ME played a key role in conceptualization, data validation, methodology. PJ contributed to data curation and review and editing of the draft. SS focused on visualization, re-drafting and editing while AS and EP made major contributions by correcting the final version of the manuscript. SKB contributed to reviewing and final proofreading of the manuscript. All authors read and approved the final manuscript.

Compliance with ethical standards

Conflict of interest: Authors do not have any conflict of interest to declare.

Ethical issues: None

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Peer review: Publisher thanks Sectional Editor and the other anonymous reviewers for their contribution to the peer review of this work.

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