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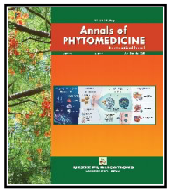

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Integrated nutrient management approaches for enhancing the phytomedicinal and quality parameters of okra (*Abelmoschus esculentus* (L.) Moench)

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Abstract

The study evaluated the influence of varying levels of farmyard manure (FYM), applied on a nitrogen equivalent basis to the recommended dose of nitrogen (RDN), in combination with three organic nutrient modules on the phytochemical quality, antioxidant potential, and anti-nutritional characteristics of okra. The experiment was conducted for two consecutive late kharif seasons (2021-22 and 2022-23) using a contrast factorial randomized block design (CFRBD) with ten treatments including a chemical-fertilizer control. Results showed that higher FYM levels (100% RDN) markedly enhanced chlorophyll content, mucilage, ascorbic acid, moisture, phenolics, and flavonoids, reflecting improved physiological activity and secondary metabolite synthesis. Among the organic modules, Module-1 comprising *Trichoderma viride* enriched FYM, neem cake, seed treatment, panchagavya, neem oil, and microbial biocontrol agents proved consistently superior. The combination L_1M_1 (100% RDN FYM + Module-1) recorded maximum phenolic content (87.60 mg GAE/100 g), flavonoids (103.75 mg QE/100 g), chlorophyll (1.19 mg/100 g), mucilage (3.87 g/100 g), and lowest IC_{50} (2.92 mg/ml), signifying strong antioxidant activity. PCA revealed that PC1 alone explained 94.67% of total variation, primarily driven by phenolics, flavonoids, moisture, and ascorbic acid. Overall, integrating full FYM with biologically enriched organic modules significantly improved okra's nutraceutical quality while minimizing antinutritional factors, demonstrating its potential as a sustainable alternative to inorganic fertilization.

1. Introduction

Abelmoschus esculentus (L.) Moench, commonly known as lady's finger or bhindi, is a nutritionally rich and economically important vegetable grown extensively in tropical and subtropical regions of Asia, Africa, and the Mediterranean region, belonging to the family Malvaceae; okra is believed to have originated in tropical Africa and has gained worldwide importance for its high nutritive value and multifaceted industrial uses (Durazzo *et al.,* 2019). The tender pods are an excellent source of carbohydrates, proteins, dietary fibre, vitamins, and essential minerals such as potassium, calcium, magnesium, and iron (Elkhalifa *et al*., 2021). The crop is also known for its high mucilage and polysaccharide content, which contribute desirable viscosity and thickening properties to processed foods such as soups, sauces, and bakery products (Petropoulos *et al.,* 2018). Fresh okra pods contain nearly 90% moisture and are rich in calcium, phosphorus, and vitamin C, along with trace elements like iron and aluminium (Habtemariam *et al*., 2019). In addition to its nutritional value, okra possesses pharmaceutical potential due to its abundance of flavonoids and biopolymers with antioxidant and antidiabetic properties (Liu *et al.,* 2019). The dried pods and seeds contain significant amounts of oil (13-22%) and protein (20-24%), making them suitable for edible oil extraction and animal-feed enrichment. Despite its nutritional and commercial importance, okra cultivation often depends heavily on chemical fertilizers and pesticides. Continuous and unbalanced use of synthetic inputs has caused soil degradation, environmental contamination, and health hazards due to residual accumulation (Kumar *et al.,* 2021). Such practices have also been associated with declining produce quality and reduced consumer acceptability. Hence, there is an urgent need to develop sustainable nutrient management strategies that maintain productivity while ensuring superior fruit quality and environmental safety.

Organic nutrient management provides a viable alternative through the use of renewable and biologically active inputs that promote nutrient recycling, microbial activity, and plant metabolic efficiency. Farmyard manure (FYM), vermicompost, panchagavya, jeevamruth, and cow-based biostimulants are key components of organic management systems that enhance nutrient availability and improve crop physiological functions (Oyege *et al.,* 2023). Recent studies have shown that organic liquid formulations and biostimulants can significantly enhance growth, yield, and quality attributes in okra and other horticultural crops (Rachamalla *et al*., 2024; Deori *et al.,* 2025). Organic nutrient management approaches combining FYM, vermicompost, and bioformulations such as *T. viride*, *Pseudomonas*

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fluorescens, vesicular arbuscular mycorrhizae (VAM) and panchagavya have proven effective in improving fruit biochemical composition, antioxidant content, and overall crop performance. Growing consumer awareness of food safety and the global emphasis on sustainable agriculture, aligned with the sustainable development goals (SDG 2: Zero hunger, SDG 12: Responsible consumption and production and SDG 13: Climate action), have accelerated the adoption of organic vegetable cultivation (Naik *et al.,* 2025). Numerous studies have examined the effects of organic and inorganic nutrient sources on okra yield and growth, limited information is available on their combined influence on biochemical quality and antioxidant characteristics, particularly under semi-arid conditions of Telangana. Investigating these relationships is essential to develop nutrient management strategies that enhance fruit quality while maintaining environmental sustainability. Therefore, the present study was undertaken to evaluate the effects of varying levels of farmyard manure (FYM) and recommended doses of fertilizers (RDF), integrated with different organic nutrient modules, on the biochemical composition, antioxidant potential, and fruit quality of okra.

2. Materials and Methods

2.1 Experimental details

The study was conducted during the late kharif seasons of 2021-22 and 2022-23 at the Post Graduate Institute of Horticultural Sciences (PGIHS), Mulugu, under Sri Konda Laxman Telangana Horticultural University, Telangana, India. The experimental site lies in a semi-arid, sub-tropical agro-climatic zone located at 17°432 023 N latitude, 78°372 343 E longitude, and an elevation of 595 m above mean sea level.The seeds of okra used in the present study were procured from the centre for sustainable agriculture (CSA), Hyderabad, representing a desi local organic variety. For authentication and future reference, a Voucher Specimen was deposited under the accession number CHR/202102 in the same Institute. Experiment was carried out under organic nutrient management systems to assess the impact of various nutrient sources and organic modules on the quality of okra. Experiment was laid out in contrast factorial randomized block design (CFRBD) with three replicationsand the means separated using least significant difference (LSD) at $p<0.05$, performed.

2.2 Experimental details

This experiment evaluated the effect of different levels of farmyard manure (FYM), applied based on nitrogen equivalence to the recommended dose of nitrogen (RDN), in combination with organic nutrient management modules.

Factor I: Levels of FYM (on N equivalent basis)

L_1 : FYM equivalent to 100% RDN

L_2 : FYM equivalent to 75% RDN

L_3 : FYM equivalent to 50% RDN

(RDF for okra = 100:50:50 kg NPK ha^{-1}; FYM applied on nitrogen equivalence)

Factor II: Organic nutrient management modules

Organic nutrient management modules

Module 1 (M_1): This module involves the application of *Trichoderma viride* at 5 kg/ha enriched with FYM and neem cake at 250 kg/ha, incorporated during the final ploughing. Seeds are treated with *T. viride* at 4 g/kg seed. Foliar spraying of 3% panchagavya and 5% neem oil is carried out at 10-day intervals until the final harvest. Additionally, *Beauveria bassiana* at 5 g/l and *Bacillus thuringiensis* at 1 kg/ha are sprayed at 10day intervals starting from flowering initiation.

Module 2 (M_2): This module includes the application of *P. fluorescens* at 5 kg/ha enriched with FYM and neem cake at 250 kg/ha, incorporated during the final ploughing. Seed treatment is done with Bacillus macerans at 3% (w/w). Foliar sprays of 10% vermiwash and 5% neem seed kernel extract (NSKE) are provided every 10 days up to the final harvest. Spraying of Metarhizium anisopliae at 5 g/l and NPV at 250 LE/ha is carried out at 10day intervals from the initiation of flowering.

Module 3 (M_3): This module consists of applying vesicular arbuscular mycorrhiza (VAM) at 10 kg/ha enriched with FYM and neem cake at 250 kg/ha during final ploughing. Seed treatment is performed using beejamruth at 10%. Foliar sprays of 10% jeevamruth and 5% neemastra are provided at 10-day intervals until the final harvest. Spraying of *Lecanicillium lecanii* at 5 g/l and a mixture of *Trichoderma + P. fluorescens* at 5 g/l is done at 10 day intervals starting from flower initiation.

A separate control plot receiving 100% RDF (100:50:50 kg NPK ha^{-1}) was maintained for comparison.

2.3 Crop establishment and management

Okra seeds (local desi variety) were sown manually at a spacing of 45 × 45 cm in plots measuring 7.5 × 5.0 m. A light irrigation was provided immediately after sowing to ensure uniform germination. The soil of the experimental field was sandy clay, well-drained, and of medium fertility with respect to available N, P, and K. All agronomic practices such as weeding, irrigation, and plant protection were carried out uniformly across treatments as per standard recommendations.

2.4 Observations and analytical procedures

Quality parameters and soil nutrient analyses were performed following standard scientific methodologies:

2.4.1 Crude fibre content (%)

Crude fibre content in okra fruits was determined following the standard method described by AOAC (1990). A representative ground fruit sample of 2 g was refluxed with 1.25% H_2SO_4, thoroughly washed, and then refluxed again with 1.25% NaOH for 30 min. The residue obtained was washed, dried, weighed, and subsequently ignited in a muffle furnace. The loss in weight due to ignition represented the crude fibre content, which was expressed as a percentage using the following formula:

$$= \frac{\text{Weight of the dried sample (g)} - \text{Weight of the ignited sample (g)}}{\text{Initia weight of the sample (g)}}$$

2.4.2 Total soluble solids (°Brix)

The total soluble solids (TSS) of okra fruits were estimated using a hand refractometer and expressed as percentage °Brix.

2.4.3 Ascorbic acid (mg 100 g^{-1})

Ascorbic acid content was determined using the 2,6-dichlorophenol indophenol visual titration method as outlined by Ranganna (1986) and expressed as mg 100 g^{-1} of fresh weight.

Preparation of solutions: 50 mg of 2, 6-dichlorophenol indophenol sodium salt and 42 mg of sodium bicarbonate were dissolved in 150 ml of hot distilled water and the volume was made up to 200 ml.30 g of metaphosphoric acid was dissolved in a small volume of distilled water and diluted to 1000 ml.

Procedure: A 10 g sample of freshly ground okra was homogenized with 3% metaphosphoric acid and the volume was made up to 50 ml with the same solution. The extract was filtered through Whatman No. 1 filter paper. A 10 ml aliquot of the filtrate was titrated against the standard dye solution to a pink endpoint. Ascorbic acid content was calculated using the formula:

$$= \frac{\text{Tite value} \times \text{Dye factor} \times \text{Volume made up} \times 100}{\text{Aliquot of extract taken for estimation} \times \text{Weight of sample taken for estimation}}$$

Dye factor = 0.5/ Average burette reading for standardization of dye solution

2.4.4 Chlorophyll content in fresh fruit (mg 100 g^{-1})

Total chlorophyll content was estimated using the method proposed by Arnon (1949). Fresh fruit samples (3 g) from the median portion were homogenized with 10 mg of magnesium sulphate and 30 ml of 80% acetone (v/v). The extract was filtered and made up to 50 ml with acetone. Absorbance was recorded spectrophotometrically at 645 nm and 663 nm, and the total chlorophyll content was computed and expressed as mg 100 g^{-1} of fresh weight.

2.4.5 Mucilage content (g 100 g^{-1})

Mucilage content was determined according to the method described by Farooq *et al.* (2013) with slight modifications. Fresh okra pods from each treatment were sliced, shade-dried for 24 h and oven-dried at 50°C for 72 h until constant weight. The dried pods were powdered, and 100 g of the powder was extracted with 300 ml of distilled water. The mixture was heated at 60°C for 1 h with continuous stirring and filtered through muslin cloth. The filtrate was treated with 20 ml of acetone and allowed to stand for 30 min before refiltration. The mucilage residue obtained was oven-dried at 45°C until a constant weight was achieved. Mucilage content was calculated using the formula:

$$= \frac{\text{Weight of dried mucilage residue (g)}}{\text{Initial weight of the sample (g)}} \times 100$$

2.4.6 Total phenolic content

The total phenolic content (TPC) of okra samples was quantified using the folin-ciocalteureagent (FCR) method as described by Duha and Yed (1997), with slight modifications. The results were expressed as milligrams of gallic acid equivalents per 100 g of dry weight (mg GAE/100 g DW), based on a calibration curve generated from gallic acid standards ($Y = 0.072x - 0.0356$; $R^2 = 0.9844$), where x represents the gallic acid concentration and Y denotes the absorbance. For the analysis, an aliquot of 0.1 ml from the fresh or treated okra extract was transferred into a 10 ml volumetric flask and diluted with 2.5 ml of distilled water. Subsequently, 250 µl of FCR was added and mixed thoroughly. After allowing the reaction to proceed for 3 min, 0.5 ml of 10% (w/v) sodium carbonate solution was introduced. The mixture was kept in the dark for 1 h to facilitate colour development, after which the absorbance was measured at 760 nm using a spectrophotometer (Model UVD-2900, Labomed, Los Angeles, USA). Each determination was carried out in triplicate to ensure accuracy and reproducibility.

2.4.7 Moisture content (%)

Tender okra pods were randomly collected from each experimental plot at harvest, immediately weighed to record the fresh weight, and then sliced into small pieces. The samples were oven-dried at 75°C for 72 h until a constant weight was obtained. The final dry weight was recorded, and the moisture content was determined from the difference between fresh and dry weights using the standard procedure described by Agle and Woodbury (1968):

$$\text{Moisture content (\%)} = \frac{\text{Fresh weight} - \text{Dry weight}}{\text{Fresh weight}} \times 100$$

2.4.8 Total flavonoid content (mg QE/100 g)

The total flavonoid content of fresh and treated okra samples was quantified using the aluminum chloride colorimetric method as described by Miliuskas *et al*. (2004), with minor modifications. For analysis, 0.2 ml of ethanolic extract from each treatment was mixed with 1 ml of aluminum trichloride solution (20 g/l) and the volume was made up to 25 ml with distilled water. The mixture was incubated for 40 min at 20°C, after which the absorbance was measured at 415 nm using a UV-Visible spectrophotometer. Quercetin Standard solutions were prepared and analyzed under the same conditions to generate a calibration curve. The total flavonoid content in each sample was expressed as milligrams equivalents per 100 g of dry weight (mg QE/100 g DW) and calculated using the regression equation derived from the calibration curve:$Y = 0.0933\ x - 0.0396$ (R^2=0.987)

where x represents the quercetin concentration and Y the absorbance. All analyses were conducted in triplicate to ensure accuracy and reproducibility.

2.4.9 Antioxidant activity (DPPH IC_{50}, mg/ml)

The antioxidant potential of okra fruit extracts was evaluated using the DPPH (1,1-diphenyl-2-picrylhydrazyl) free radical scavenging assay following the procedure described by Turkmen *et al.* (2005), with slight modifications. Ethanolic extracts were prepared at varying concentrations (10 g/100 ml), and 1 ml of each extract was mixed with 1 ml of DPPH methanolic solution (6×10^{-1} M). The mixture was incubated for 30 min in the dark at room temperature to allow for reaction stabilization. After incubation, the absorbance was recorded at 517 nm using a UV-Visible spectrophotometer against a blank. Butylated hydroxytoluene (BHT, 200 ppm) was used as a positive control, and methanol served as the reagent blank. The percentage of DPPH radical scavenging activity was calculated using the following equation:

$$\text{DPPH scavenging activity (\%)} = \frac{\text{A control} - \text{A sample}}{\text{A control}} \times 100$$

The IC_{50} value (the concentration of extract required to scavenge 50% of DPPH radicals) was derived from the plotted graph of percentage inhibition versus extract concentration. Lower IC_{50} values indicate stronger antioxidant activity. All analyses were conducted in triplicate to ensure precision.

2.4.10 Oxalate content

The oxalate concentration in okra fruits was estimated using the standard method described by the Association of Official Analytical Chemists (AOAC, 2005), with minor modifications. One gram of finely ground okra sample was placed in a 100 ml conical flask, and 75 ml of 3M sulfuric acid (H_2SO_4) was added while continuously stirring on a magnetic stirrer for one hour to facilitate extraction. The mixture was then filtered through Whatman No. 42 filter paper. A 25 ml aliquot of the filtrate was titrated against 0.1N potassium permanganate ($KMnO_3$) solution while heating on a hotplate maintained at 80-90 °C until a faint pink color persisted, indicating the end point. The oxalate content was calculated based on the titration value and expressed in milligrams per 100 g of sample.

2.4.11 Tannin content

Tannin content was determined colorimetrically following the method of Joslyn (1970). About 0.5 g of finely ground okra flour was defatted using 5% ethyl ether for at least 15 min to remove interfering lipids. The defatted residue was then extracted with methanol to obtain tannin-rich extracts. The absorbance of the resulting solution was measured at 760 nm using a UV-Visible spectrophotometer. The tannin concentration was quantified using a calibration curve constructed from standard tannic acid solutions and expressed as milligrams per 100 g of sample.

2.4.12 Principal component analysis (PCA)

Principal component analysis (PCA) was performed to identify the main sources of variation and interrelationships among twelve biochemical and quality traits, including total phenolic content, total flavonoid content, DPPH IC_{50}, tannins, oxalates, moisture, mucilage, TSS, ascorbic acid, chlorophyll, and crude fibre, across all ten treatments (L_1M_1 - L_3M_3 and Control). The standardized mean data of these parameters were subjected to PCA using the correlation matrix method to reduce data dimensionality and to visualize treatment clustering. The eigen values, proportion of variance and component loadings were computed to determine the contribution of each trait to total variability. Biplots were generated to illustrate the distribution and grouping of treatments along the first two principal components. All analyses were carried out using R software (version 4.3.1).

3. Results

3.1 Crude fibre content (%)

Crude fibre content in okra fruits increased significantly with higher levels of FYM, indicating that nutrient enrichment from organic manure enhanced fibre synthesis through improved vegetative growth and lignification. The lowest crude fibre (7.25%) was recorded at L_1, while the highest (9.25%) occurred at L_3 . Among the organic modules, M_1(8.46%) showed slightly higher fibre content than M_1 (8.20%) and M_3(8.33%), though the differences were marginal. The interaction between FYM levels and organic modules was non-significant, suggesting that FYM levels predominantly influenced fibre accumulation. The control (8.72%) showed intermediate values, confirming the beneficial role of FYM in improving pod texture and physiological quality.The minimum crude fibre content (Table 1) was observed with L_1 M_1 (FYM equivalent to 100% RDN + Organic Module-1).

3.2 Total soluble solids (TSS)

Total soluble solids (TSS) content in okra fruits decreased significantly with increasing levels of FYM, indicating a dilution effect resulting from enhanced vegetative growth and higher moisture content at elevated FYM levels. The highest TSS (7.19 °Brix) was recorded at the lowest FYM level (L_1), while the lowest (6.12 °Brix) occurred at L_3. Among the organic modules, M_1 (6.83 °Brix) showed slightly higher TSS than M_1 (6.51 °Brix) and M_3 (6.73 °Brix). The interaction between FYM levels and organic modules was non-significant, suggesting that FYM levels independently influenced soluble solids accumulation. The control (6.90 °Brix) recorded moderate values, confirming that organic nutrient sources, while promoting plant growth, slightly reduced TSS concentration compared to lower FYM applications.

Table 1: Effect of different levels of farm yard manure and organic modules on crude fibre (%) and TSS in fruits of okra

FYM levels	Crude fibre (%) Organic modules				TSS (°Brix) Organic modules			
	M_1	M_2	M_3	Mean	M_1	M_2	M_3	Mean
L_1	7.10	7.39	7.27	7.25	7.33	7.06	7.19	7.19
L_2	8.38	8.59	8.46	8.48	6.90	6.58	6.78	6.75
L_3	9.11	9.41	9.25	9.25	6.26	5.89	6.22	6.12
Mean	8.20	8.46	8.33		6.83	6.51	6.73	
Control			8.72				6.90	
	L	M	L × M	Control	L	M	L × M	Control
SEm±	0.02	0.02	0.04	0.04	0.02	0.02	0.05	0.05
LSD (5%)	0.04	0.04	NS	0.16	0.05	0.05	NS	0.19

Factor: 1

L_1: Farm yard manure equivalent to 100% RDN

L_2: Farm yard manure equivalent to 75% RDN

L_3: Farm yard manure equivalent to 50% RDN

Factor: 2

M_1: Organic module-1

M_2:Organic module-2

M_3: Organic module-3

Control: 100% Recommended dose of fertilizers

3.3 Chlorophyll content (mg/100 g)

Chlorophyll content in okra fruits decreased significantly with the reduction in FYM levels, highlighting the crucial role of organic nutrient supply in sustaining pigment synthesis and photosynthetic efficiency (Table 2). The highest chlorophyll content (1.13 mg/100 g) was recorded at L_1 (FYM equivalent to 100% RDN), whereas the lowest (0.69 mg/100 g) was observed at L_3 (FYM equivalent to 50% RDN). Among the organic modules, M_3 (0.95 mg/100 g) recorded higher chlorophyll content compared to M, (0.83 mg/100 g) and M_3 (0.90 mg/100 g), though differences were marginal. The interaction between FYM and organic modules was non-significant, indicating that both factors influenced chlorophyll accumulation independently. The control treatment (0.98 mg/100 g) recorded intermediate chlorophyll content, confirming that the application of FYM and organic modules sustained pigment stability better than inorganic fertilizers.

3.4 Mucilage content (g/100 g)

Mucilage content in okra fruits exhibited a significant declining trend with decreasing FYM levels, highlighting the pivotal role of organic nutrient inputs in stimulating polysaccharide biosynthesis. The highest mucilage content (3.71 g/100 g) was recorded at L_1(FYM equivalent to 100% RDN), followed by L_1(3.31 g/100 g) and L*f* (3.00 g/100 g). Among the organic modules, M_2 (3.45 g/100 g) consistently outperformed M, (3.23 g/100 g) and M_3 (3.34 g/100 g). The control (3.12 g/100 g) exhibited comparatively lower mucilage content, emphasizing that integrated organic nutrient management enhanced qualitative fruit attributes more effectively than inorganic fertilization.

3.5 Ascorbic acid content (mg/100 g)

Ascorbic acid content in okra fruits showed a significant decreasing trend with the reduction in FYM levels, indicating that organic nutrient enrichment plays a crucial role in promoting antioxidant accumulation. The highest ascorbic acid concentration (15.19 mg/100 g) was recorded at L_1 (FYM equivalent to 100% RDN), whereas the lowest (12.26 mg/100 g) was observed at L_3(FYM equivalent to 50% RDN). Among the organic modules, M_1 (14.08 mg/100 g) exhibited the highest ascorbic acid content, followed by M_3 (13.66 mg/100 g) and M_3 (13.16 mg/100 g). The control treatment (14.40 mg/100 g) recorded intermediate values. The non-significant interaction (L × M) suggested that FYM levels independently influenced ascorbic acid accumulation. The treatment L_1 M_2 (FYM equivalent to 100% RDN + Organic module-1) consistently recorded the maximum ascorbic acid content in compare to control (Table 2).

3.6 Total phenolic content (mg GAE/100 g)

Total phenolic content (TPC) was significantly influenced by both FYM levels and organic modules, with a distinct interaction effect ($L_1 \times M_1$). The highest TPC (87.60 mg GAE/100 g) occurred under L_1M_1 (100 % RDN FYM + Module-1), followed by L_1M_3 (84.23 mg) and L_1M_2 (81.60 mg) (Table 3). The lowest (74.60 mg) was observed in L_3M_2 (50% RDN + Module-2). The control (81.80 mg GAE/100 g) exhibited moderate TPC, lower than L_1M_1 but comparable to L_2 treatments, showing that well-balanced organic inputs can match or surpass chemical fertilization.

3.7 Moisture content (%)

Moisture content (%) was positively affected by both FYM level and organic module, showing a mild but meaningful $L_1 \times M_1$ interaction. The maximum moisture (92.03%) was obtained with L_1M_1 (100% RDN FYM + Module-1), followed by L_1M_2 (90.80%) and control (90.50%) (Table 3). The lowest moisture content (86.00%) was recorded in L_3M_2 (50% RDN FYM + Module-2). The trend reveals that higher FYM and well-structured organic modules improve soil moisture retention, plant hydration, and consequently tissue water content.

3.8 Total flavonoid content (mg QE/100 g)

Total flavonoid content (TFC) exhibited a strong positive response to both FYM levels and organic modules, with a significant $L_1 \times M_1$ interaction. The highest TFC (103.75 mg QE/100 g) occurred under L_1M_1 (100% RDN + Module-1), followed by L_1M_2 (101.55 mg) and L_1M_3 (101.93 mg). The lowest value (92.69 mg) was noted in L_3M_2 (50 % RDN + Module-2). The control (98.95 mg QE/100 g) (Table 3) exhibited lower flavonoid content than L_1 treatments but was comparable to L_2, indicating that chemical fertilization maintains moderate antioxidant levels, though not as high as organic enrichment.

3.9 Antioxidant activity (DPPH IC_{50} mg/ml)

The antioxidant potential, expressed as DPPH IC_{50} (mg/ml), varied significantly across FYM levels, organic modules, and their interaction. The lowest IC_{50} value, indicating the highest antioxidant activity, was recorded in L_1M_1 (2.92 mg/ml), while the highest (3.80 mg/ml) was obtained in L_3M_2 (Table 4). The mean IC_{50} increased from L_1 (3.02 mg/ml) to L_3 (3.73 mg/ml), demonstrating that antioxidant efficiency declined with decreasing organic nutrient levels. Among the organic modules, M_1 (3.26 mg/ml) showed superior antioxidant capacity compared to M_2 (3.40 mg/ml) and M_3 (3.38 mg/ml).The interaction clearly indicates a synergistic influence, where 100% RDN FYM with biologically enriched M_1 enhanced antioxidant enzymes and radical scavenging activity. The control (3.30 mg/ml) exhibited moderate antioxidant activity, similar to L_2 treatments, but was inferior to L_1M_1, confirming that organic nutrient management fosters higher antioxidant potential.

3.10 Tannin content (mg/100 g)

Tannin concentration increased significantly with decreasing FYM levels, showing distinct variation among organic modules and their interaction (L × M). The highest tannin content (5.68 mg/100 g) was recorded under L_3M_2 (50% RDN + Module-2), whereas the lowest (4.75 mg/100 g) occurred in L_1M_1 (100% RDN + Module-1) (Table 4). Mean tannin values across FYM levels increased from 4.90 (L_1) to 5.58 (L_3), suggesting that nutrient stress at lower FYM levels stimulated tannin biosynthesis. Among modules, M_2 (5.37 mg/100 g) consistently produced higher tannins than M_1 (5.13 mg/100 g) and M_3 (5.26 mg/100 g). The interaction revealed that mild nutrient limitation may trigger phenolic polymerization, resulting in increased tannin accumulation. However, higher tannins may negatively influence sensory quality due to astringency. The control (5.27 mg/100 g) showed intermediate tannin levels, close to L_2M_3.

3.11 Oxalate content (mg/100 g)

Oxalate accumulation varied significantly with FYM levels and organic modules, though the $L_1 \times M_2$ interaction was statistically nonsignificant. The highest oxalate content (0.70 mg/100 g) was

observed in L_3M_2 (50% RDN + Module-2), while the lowest (0.40 mg/100 g) occurred under L_1M_1 (100% RDN + Module-1) (Table 4). Mean oxalate values increased progressively from L_1 (0.44 mg/100 g) to L_3 (0.66 mg/100 g), indicating that reduced FYM supply stimulated oxalate synthesis. Organic modules influenced oxalate content marginally, with M_2 (0.59 mg/100 g) showing slightly higher accumulation compared to M_1 (0.51 mg/100 g). The control (0.53 mg/100 g) exhibited oxalate concentration comparable to L_2M_1, suggesting that balanced chemical fertilization maintains moderate oxalate levels.

Table 2: Effect of different levels of farm yard manure and organic modules on chlorophyll (mg/100 g), mucilage (g/100 g) and ascorbic acid (mg/100 g) content in fruits of okra

FYM levels	Chlorophyll (mg/100 g)				Mucilage (g/100 g)				Ascorbic acid (mg/100 g)			
	Organic modules				Organic modules				Organic modules			
	M_1	M_2	M_3	Mean	M_1	M_2	M_3	Mean	M_1	M_2	M_3	Mean
L_1	1.19	1.07	1.15	1.13	3.87	3.58	3.69	3.71	15.69	14.59	15.30	15.19
L_2	0.93	0.81	0.86	0.87	3.39	3.23	3.31	3.31	13.88	13.02	13.45	13.45
L_3	0.75	0.61	0.71	0.69	3.09	2.89	3.02	3.00	12.68	11.88	12.23	12.26
Mean	0.95	0.83	0.90		3.45	3.23	3.34		14.08	13.16	13.66	
Control			0.98			3.12					14.40	
	L	M	L × M	Control	L	M	L × M	Control	L	M	L × M	Control
SEm±	0.004	0.004	0.011	0.011	0.02	0.02	0.04	0.04	0.02	0.02	0.06	0.06
LSD (5%)	0.011	0.011	NS	0.043	0.04	0.04	0.03	0.20	0.06	0.06	NS	0.26

Factor: 1

L_1: Farm yard manure equivalent to 100% RDN

L_2: Farm yard manure equivalent to 75% RDN

L_3: Farm yard manure equivalent to 50% RDN

Factor: 2

M_1: Organic module-1

M_2:Organic module-2

M_3: Organic module-3

Control: 100% Recommended dose of fertilizers

Table 3: Effect of different levels of farm yard manure and organic modules on total phenolic content (mg GAE/100 g FW), moisture content (%) and total flavonoid content (mg QE/100 g) content in fruits of okra

FYM levels	Total phenolic content (mg GAE/100 g FW)				Moisture content (%)				Total flavonoid content (mg QE/100g)			
	Organic modules				Organic modules				Organic modules			
	M_1	M_2	M_3	Mean	M_1	M_2	M_3	Mean	M_1	M_2	M_3	Mean
L_1	87.60	81.60	84.23	84.48	92.03	90.80	91.20	91.34	103.75	101.55	101.93	102.41
L_2	79.97	78.93	80.00	79.63	91.00	90.20	89.83	90.34	99.81	98.14	97.66	98.54
L_3	78.63	74.60	77.27	76.83	87.50	86.00	86.63	86.71	94.65	92.69	93.56	93.63
Mean	82.07	78.38	80.50		90.18	89.00	89.22		99.40	97.46	97.72	
Control			81.80				90.50			98.95		
	L	M	L × M	Control	L	M	L × M	Control	L	M	L × M	Control
SEm ±	0.06	0.06	0.17	0.17	0.02	0.02	0.05	0.05	0.02	0.02	0.06	0.06
LSD (5%)	0.17	0.17	0.51	0.66	0.05	0.05	0.16	0.18	0.06	0.06	0.19	0.25

Factor: 1

L_1: Farm yard manure equivalent to 100% RDN

L_2: Farm yard manure equivalent to 75% RDN

L_3: Farm yard manure equivalent to 50% RDN

Factor: 2

M_1: Organic module-1

M_2:Organic module-2

M_3: Organic module-3

Control: 100% Recommended dose of fertilizers

3.12 Principal component analysis (PCA)

Principal component analysis(PCA) was performed to examine the multivariate relationships among eleven biochemical and nutritional traits of the treatments (L_1M_1-L_3M_3 and Control). PCA using the correlation matrix revealed that the first principal component (PC1) explained 94.67% of the total variation, while PC2 accounted for 2.43% (Figure 1). Together, PC1 and PC2 captured over 97% of the cumulative variation, indicating that the majority of differences among treatments could be summarized along these two axes. The dominance of PC1 suggests that most of the variation among treatments is strongly associated with a combination of traits that contribute

positively or negatively to this component. Traits such as total phenolic content, total flavonoid content, ascorbic acid, and moisture content had higher loadings on PC1, indicating that these parameters were the primary drivers of variability among the treatments. In contrast, PC2 captured minor variations, which may reflect subtle differences in secondary traits such as crude fibre, chlorophyll content, tannins, contributingto treatment separation. The PCA biplot (PC1 vs PC2) revealed clear clustering of treatments. Treatments under the L_1 group (L_1M_1-L_1M_3) were positioned on the positive side of PC1, indicating higher levels of phenolics, flavonoids, and antioxidant activities, whereas the L_3 treatments (L_3M_1-L_3M_3) clustered on the negative side of PC1, reflecting lower nutrient and bioactive compound content. The Control treatment was intermediate, positioned near the centre, suggesting moderate levels of the analysed traits. This separation demonstrates that the combination of different levels of treatment factors significantly influenced the biochemical and nutritional profiles of the samples. The strong association of PC1 with key antioxidant and nutrient-related traits aligns with previous studies reporting that phenolic compounds, flavonoids, and ascorbic acid contribute substantially to the bioactive potential and nutritional quality of plant-derived foods. The results indicate that treatments with higher PC1 scores (L_1 group) possess enhanced functional properties, including antioxidant potential and nutrient density, which could be valuable for both health-promoting effects and industrial applications.

Table 4: Effect of different levels of farm yard manure and organic modules on antioxidant activities DPPH IC_{50} (mg/ml), tannins (mg/100 g) and oxalates (mg/100 g) content in fruits of okra

FYM levels	Antioxidant activities DPPH IC_{50} (mg/ml)				Tannins (mg/100 g)				Oxalates (mg/100 g)			
	Organic modules				Organic modules				Organic modules			
	M_1	M_2	M_3	Mean	M_1	M_2	M_3	Mean	M_1	M_2	M_3	Mean
L_1	2.92	3.03	3.10	3.02	4.75	5.05	4.90	4.90	0.40	0.47	0.44	0.44
L_2	3.20	3.37	3.32	3.30	5.18	5.37	5.28	5.28	0.52	0.59	0.56	0.56
L_3	3.65	3.80	3.72	3.73	5.46	5.68	5.59	5.58	0.61	0.70	0.67	0.66
Mean	3.26	3.40	3.38		5.13	5.37	5.26		0.51	0.59	0.56	
Control			3.30				5.27				0.53	
	L	M	L × M	Control	L	M	L × M	Control	L	M	L × M	Control
Sem±	0.01	0.01	0.02	0.02	0.02	0.02	0.04	0.04	0.01	0.01	0.03	0.03
LSD (5%)	0.02	0.02	0.50	0.50	0.04	0.04	0.50	0.60	0.03	0.03	NS	0.04

Factor: 1

L_1: Farm yard manure equivalent to 100% RDN

L_2: Farm yard manure equivalent to 75% RDN

L_3: Farm yard manure equivalent to 50% RDN

Factor: 2

M_1: Organic module-1

M_2:Organic module-2

M_3: Organic module-3

Control: 100% Recommended dose of fertilizers

Table 5: Principal component analysis (PCA) summary of variance explained by ten components

Component	Standard deviation	Proportion of variance	Cumulative proportion
PC1	3.2270	0.9467	0.9467
PC2	0.51718	0.02432	0.97102
PC3	0.42946	0.01677	0.98779
PC4	0.29725	0.00803	0.99582
PC5	0.12982	0.00153	0.99735
PC6	0.11291	0.00116	0.99851
PC7	0.09775	0.00087	0.99938
PC8	0.0664	0.00040	0.99978
PC9	0.04900	0.00022	1.00000
PC10	1.187e-15	0.00000	1.00000

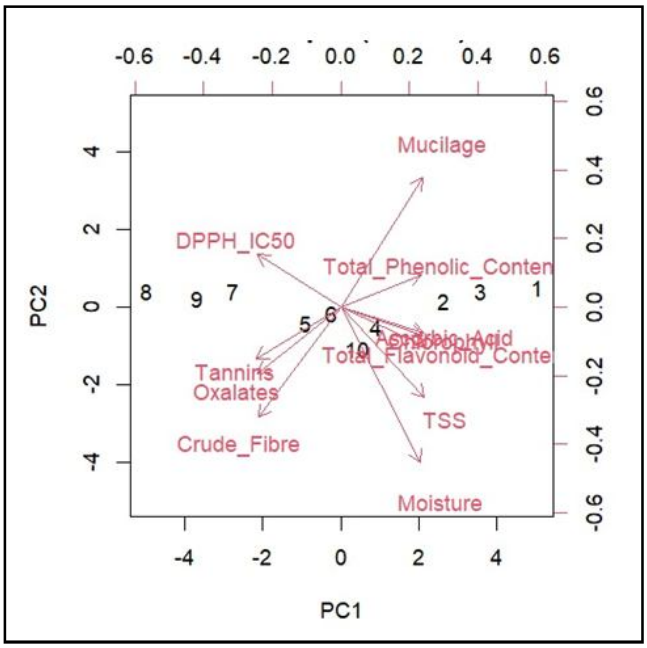

Figure 1: PCA biplot of effect of different levels of FYM and organic modules on phytomedicinal and quality parameters

4. Discussion

The application of organic manure (FYM equivalent to 100% RDN + Organic Module-1) demonstrated a clear superiority in enhancing okra fruit quality compared to inorganic fertilizers, primarily due to a balanced C:N ratio and improved nutrient synchronization that enhanced physiological functioning. Enhanced crude fibre content with advancing maturity may be attributed to reduced tissue succulence caused by increased cell wall lignification and limited nitrogen uptake at later growth stages (Alam *et al*., 2019). Similar reductions in crude fibre with FYM application were recorded by Premsekhar and Rajashree (2009), while foliar application of panchagavya @ 3% also reported minimum crude fibre levels (Hathi *et al*., 2022). These findings corroborate earlier reports by Amiry*et al*. (2018), and Alam *et al*. (2019) supporting the role of organic nutrient sources in quality enhancement. Higher TSS values under treatment L_1 M_2 may be associated with improved nutrient availability enhancing photosynthate production and carbohydrate accumulation. Nitrogen promotes biomass and assimilates synthesis, while potassium regulates carbohydrate metabolism and conversion of starch to sugars *via* sucrose synthetase activation. Similar observations were reported by Chassy *et al*. (2006) in tomatoes under organic management and by Arivazhagan *et al.* (2019) who recorded significant improvement in TSS and ascorbic acid with organic manures and panchagavya. Comparable enhancements in biochemical quality were also observed by Shida *et al.* (2020) in Chinese cabbage. Improved chlorophyll content under integrated organic application may be attributed to adequate supply of macro and micronutrients from FYM enriched with *T. viride*, neem cake, and foliar panchagavya. Nitrogen and magnesium are essential for chlorophyll synthesis, while iron and sulfur regulate chloroplast integrity and photosynthetic functioning (Suresh *et al*., 2021). Increased chlorophyll concentration is likely linked to enhanced photosynthetic activity and pigment stability (Karanatsidis and Berova, 2009).

The increase in mucilage content under higher FYM levels may relate to improved nutrient uptake, soil moisture retention, and metabolic activity supporting polysaccharide synthesis. Similar observations were reported by Sanni and Adesina (2012) and by Mageshen and Bagavathi (2022). Enhanced ascorbic acid accumulation under organic management may be due to improved carbohydrate metabolism and balanced nutrient supply. Higher potassium availability may reduce enzymatic oxidation of ascorbic acid (Mohit *et al.,* 2019). Comparable improvements were noted by Sable *et al.* (2007) in tomato and Meena *et al*. (2019) in okra, while Arivazhagan *et al*. (2019) emphasized the synergistic role of foliar panchagavya.

The highest phenolic content under L_1M_1 suggests synergistic interaction where nutrient sufficiency and microbial enhancement stimulate phenylpropanoid biosynthesis, particularly phenylalanine ammonia-lyase (PAL) activity (Ibrahim *et al*., 2013). Similar positive effects were reported by Ozdemir *et al.* (2018) in grapes and Saikia and Upadhyaya (2011) in *Asparagus racemosus*. Lower FYM levels correspondingly reduced phenolic accumulation.

Improved soil moisture status under FYM may result from enhanced porosity and water-holding capacity (Akhtar *et al*., 2023; Kumar *et al*., 2022). Slightly higher moisture under L_1M_1 reflects nutrient microbial synergy supporting turgor and postharvest freshness.

Higher flavonoid content under L_1M_1 likely originates from enhanced activation of phenylpropanoid pathways and microbial nutrient mineralization. Comparable findings were reported by Ozdemir *et al.* (2018) and Ibrahim *et al.* (2013). Elevated antioxidant activity under organic nutrition aligns with increased phenolics and flavonoids contributing to free-radical neutralization (Olawole*et al.,* 2022).

Lower tannin content under balanced nutrition suggests that chemical fertilizers sustain moderate phenolic condensation, while organic synergy better regulates secondary metabolism. These trends align with Ibrahim *et al*. (2013) and Kumar *et al*. (2022) indicating that nutrient stress elevates tannin accumulation.Oxalate reduction under L_1M_1 may be due to balanced calcium and nitrogen uptake and reduced oxalic acid formation (Libert and Franceschi, 1987; Noonan and Savage, 1999). Greater microbial activity enhances mineral chelation and nutrient balance, minimizing antinutritional accumulation.

5. Conclusion

The findings clearly demonstrate that integrating FYM at 100% RDN with biologically enriched organic modules, particularly Module-1, substantially enhances the biochemical, phytomedicinal, and nutritional quality of okra under semi-arid conditions. This combination improved key quality attributes including phenolics, flavonoids, mucilage, ascorbic acid, chlorophyll, and antioxidant activity while simultaneously reducing undesirable antinutritional factors such as oxalates and tannins. Treatments receiving lower FYM levels showed reduced nutrient uptake and weaker antioxidant profiles, confirming the importance of adequate organic nutrient supply. PCA further validated that full FYM integration strongly influenced the most valuable quality traits. Overall, the L_1M_1 treatment emerged as the most effective strategy for producing nutrient-dense, residue-free okra, offering a highly sustainable and eco-friendly alternative to conventional fertilizer-based cultivation systems.

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Conflict of interest

The authors declare no conflicts of interest relevant to this article.

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