



RESEARCH ARTICLE

Nutrient alchemy: Optimizing multicut fodder sorghum for yield, quality and environmental balance

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Abstract

This study delves into the transformative impact of diverse nutrient management practices on the yield, quality, soil health and carbon sequestration potential of multi-cut fodder sorghum. Employing a randomized block design with three replications across 11 treatments, the research evaluated the effects of organic, inorganic and biofertilizer-based nutrient applications. The findings were striking: a combination of poultry manure at 75 % nitrogen equivalent with biofertilizers (*Arbuscular mycorrhiza* and Azophos) led to a remarkable 31.2 % boost in fodder yield and a 36.4 % increase in dry matter production compared to the control. Soil analysis revealed an 8.22 % decrease in pH, alongside substantial enhancements in key nutrients, with available nitrogen, phosphorus and potassium levels soaring by 28.40 %, 44.19 % and 9.43 % respectively, under the same treatment. These practices also supercharged soil enzyme activities, with notable increases in amylase (1.21 mg reducing sugars g⁻¹ soil 24 h⁻¹), invertase (1.45 mg reducing sugars g⁻¹ soil 24 h⁻¹), cellulase (34.8 µg reducing sugars g⁻¹ soil 24 h⁻¹), phosphatase (19.73 µg phenols g⁻¹ soil 24 h⁻¹), dehydrogenase (11.56 µg triphenyl formazan g⁻¹ soil 24 h⁻¹) and urease (17.92 mg NH₄ - N g⁻¹ soil 24 h⁻¹), signaling a vibrant uptick in microbial activity and overall soil health. Moreover, this nutrient strategy delivered a 39.22 % rise in soil organic carbon (SOC) and a 38.74 % boost in soil carbon stock, highlighting its powerful potential for long-term carbon sequestration. This study offers critical insights for crafting sustainable agricultural practices that not only maximize crop production but also enhance soil fertility and contribute meaningfully to environmental conservation.

Keywords

biofertilizers; carbon sequestration; fodder quality; nutrient source; microbial activity; multicut fodder sorghum

Introduction

Sorghum is a versatile crop widely cultivated for its adaptability to diverse agro-climatic conditions and its significant contributions to food, fodder and biofuel production (1). Among its various types, multicut fodder sorghum stands out as particularly crucial for livestock feed,

providing essential nutrients and playing a key role in sustainable agricultural systems. However, optimizing the yield and quality of fodder sorghum presents challenges due to varying soil fertility, climate conditions and management practices (2). Multicut sorghum is capable of producing high-quality forage in mid to late summer when cool season perennials have low production (3). Being an exhaustive crop, sorghum's yield and quality suffer significantly if proper amounts of fertilizers are not applied. Nitrogen fertilizer, for instance, enhances the production of forage sorghum with better nutritive value, increasing crude protein, metabolizable energy, succulence and palatability of fodders (4). Additionally, phosphorus is crucial for sorghum's root growth, making it the second most deficient yet essential plant nutrient after nitrogen (5). Optimum phosphorus application rates are vital for improving crop yields (6). Biofertilizers, which are natural products carrying living microorganisms derived from the root or cultivated soil, have no ill effects on soil health and the environment. They improve the quantitative and qualitative features of many plants (7). Nutrient management is a critical factor influencing the productivity and nutritional value of fodder crops. Traditional practices often rely heavily on chemical fertilizers, which can lead to soil degradation and environmental concerns over time. Recently, there has been growing interest in integrating organic amendments, such as poultry manure and farmyard manure, with biofertilizers like *Azophos* and *Arbuscular mycorrhiza* to enhance soil health and crop performance. *Azophos* biofertilizer includes *Azospirillum* and *Phosphobacteria*, which assist plants in acquiring nitrogen and phosphorus. Microorganisms that solubilize phosphorus (P) and potassium (K) convert unavailable forms of these nutrients into soluble forms through dissolution and/or mineralization processes. Additionally, microbial metabolic activities influence soil pH, density and porosity (8, 9).

These organic and biological inputs not only provide essential nutrients but also improve soil structure, microbial activity and nutrient cycling, thereby promoting sustainable agricultural practices (10). Moreover, these practices can enhance carbon sequestration in soils, capturing atmospheric carbon dioxide and storing it as organic carbon within the soil matrix. This process not only mitigates climate change but also improves soil health and fertility. This study aims to evaluate the impact of diverse nutrient management practices on the yield, quality, soil health and carbon sequestration potential of multicut fodder sorghum. By comparing the effects of organic amendments, biofertilizers and conventional fertilizers, we seek to identify optimal strategies that enhance fodder production while maintaining soil fertility and

ecosystem health. The findings from this research will contribute to developing sustainable nutrient management practices that support high-yielding and nutritionally rich fodder sorghum, ultimately benefiting both farmers and the environment.

Materials and Methods

Experimental site

A study was conducted from December 2022 to May 2024 at Tamil Nadu Agricultural University in Coimbatore, Tamil Nadu, India to evaluate the effects of various nutrient management practices on yield, quality and soil health of multi-cut fodder sorghum. The experimental site was situated at 11°07'3.36" N latitude and 76°59'39.91" E longitude, with an elevation of 426 m above sea level. The area receives an average annual rainfall of 746.5 mm over 47 rainy days, with average annual maximum and minimum temperatures of 31.8 °C and 21.4 °C respectively.

Treatments and Experimental Setup

Eleven treatments were used in a randomized block design (RBD) with 3 replications in the experiment. The field treatments were : T₁: recommended dose of fertilizer (RDF); T₂: recommendation based on soil test values; T₃: T₂ with *Arbuscular mycorrhiza*; T₄: T₂ with *Azophos*; T₅: T₂ with both *Azophos* and *Arbuscular mycorrhiza*; T₆: recommendation based on soil test (75 % N + 100 % P₂O₅ + 75 % K₂O) with *Azophos* and *Arbuscular mycorrhiza*, T₇ is farmyard manure (FYM) at 100 % N equivalent; T₈ is poultry manure (PM) at 100 % N equivalent; T₉ is FYM at 75 % N equivalent with *Azophos* and *Arbuscular mycorrhiza*; T₁₀ is PM at 75 % N equivalent with *Azophos* and *Arbuscular mycorrhiza* finally T₁₁ is absolute control.

Preparation of FYM and PM

For field application, well-decomposed FYM aged 6 months and 8 weeks old poultry manure were used to provide sufficient nutrients and prevent potential plant damage from raw materials. Farmyard manure was prepared using a mixture of cattle dung, urine, farm biomass residues and straw, while poultry manure was produced by combining poultry droppings with bedding materials such as straw, sawdust.

Manure, bio fertilizer and Fertilizer application

The prescribed basal fertilizer amounts for multicut fodder sorghum CO (FS) 29 were 45 kg of nitrogen (N), 40 kg of phosphorus (P₂O₅) and 40 kg of potassium (K₂O)/ha. Additionally, 45 kg of nitrogen per cut (totally 225 kg for 5 cuts) was applied for the treatment with the recommended fertilizer dose (T₁). The soil test-based recommendation treatments (T₂) included full doses of nitrogen (100 % N) and phosphorus (100 % P₂O₅) and 75 % of the potassium (K₂O) dose. Treatments receiving biofertilizers involved applying *Arbuscular mycorrhiza* at 2000 g ha⁻¹ and *Azophos* at 4000 g ha⁻¹ during the initial stage. Treatments involving organic manures, such as poultry manure and farmyard manure, were applied based

Amount of organic manures need based on N equivalent =
$$\frac{\text{Nitrogen required (Kg/ha)}}{\text{N Content in organic manures (\%)}}$$
 on their nitrogen

equivalent at basal. Poultry manure, with a nitrogen content of 2.30 %, was applied at a rate of 9,740 kg/ha whereas farmyard manure, which has a nitrogen content of 0.71 %, applied at 31690 kg/ha. The quantity of organic manures required, based on nitrogen equivalence, was determined using the following equation.

Chemical characterization of manures and soil

The manures and soil samples were evaluated for various properties such as pH, electrical conductivity (EC), total organic carbon (TOC) and total nitrogen (N), phosphorus (P) and potassium (K) using established protocols. The pH and EC were measured using a pH meter and an EC meter respectively, with a 1:2.5 manure-to-water ratio (11). TOC in the manures was determined by the dry combustion method employing a muffle furnace. The macronutrient content (N, P and K) was analyzed following standard procedures (12).

Enzyme activity assays and microbial load assessment

The process described, soil was extracted and incubated with a starch substrate and enzyme activity was quantified based on the starch hydrolysis, which was used to measure the amylase activity in the soil samples (13). Soil invertase activity was determined using the method outlined by (14) and expressed as the amount of reducing sugars released day⁻¹. Cellulase activity in the soil samples was assessed by following the methodology (15), with its activity expressed as the amount of glucose released (µg) per day from the soil. Dehydrogenase activity was determined using the 2-3-5- triphenyl tetrazolium chloride reduction technique (16). Phosphatase activity was measured using p-nitrophenyl phosphate as the substrate (17). All enzyme assays reported as grams of oven-dried soil per 24 h incubation. The assessment of soil microbes was conducted according to the methods (18), which include collecting and preparing soil samples, followed by the enumeration of microbial populations using microbiological procedures.

Soil carbon fractions and balance

The process described comprises the soil extracting with distilled water, filtering the resultant solution and measuring the water-soluble carbon using an appropriate analytical method (19, 20). A modified procedure based on the method (21), which entails extracting soil samples using a particular reagent and then analysing the samples to determine the quantity of oxidizable organic carbon (22). Initially, soil organic carbon (SOC) was assessed using a wet digestion method, with a follow-up assessment after one year.

Passive carbon (mg/kg) = TOC - {Water soluble carbon (mg/kg) + Labile carbon (mg/kg)}

Permanent soil carbon stock (t ha⁻¹ year⁻¹) = Passive carbon (%) * Bulk density (mg m⁻³) * Depth (m)

Added SOC in passive carbon pool (t ha⁻¹ year⁻¹) = {Final (after one year) permanent soil carbon stock in treatments (t ha⁻¹ year⁻¹) - Initial permanent soil carbon stock (t/ha/year)}

Soil carbon stock

The methodology described, entails the following equation (23)

Soil C stock (t ha⁻¹) = TOC * BD * D

TOC - Total organic carbon (TOC %); BD - Bulk density (Mg m⁻³); D - Soil depth (cm)

Determination of green fodder yield and quality

Green fodder yield was initially measured at 80 days after planting, with subsequent measurements taken every 45-50 days. Throughout the experiment, 5 cuts were taken and the cumulative average yield data was calculated to assess the impact of various soil management practices on green fodder yield. Ten randomly selected plants from each plot were chopped using a fodder cutter and thoroughly mixed at the final harvest. After determining the sample's fresh weight, 500 g samples from each batch were obtained and dried at 70 °C in an oven until a consistent dry weight was reached. An electronic balance was used to determine the samples' dry weight, from which the dry matter content was computed. The conventional methods were used to calculate the percentages of crude protein (%) and crude fiber (%) (24).

Statistical analysis

The data from 3 replications of 11 treatments in the field experiment were analyzed statistically using analysis of variance (ANOVA), based on the methodology described in reference (25). Critical differences were computed for treatments exhibiting significant differences at the 5 % probability level, while treatments lacking significant differences were denoted as NS (non-significant).

Results

Initial soil characteristics and fertility assessment

The initial soil at the experimental site was characterized as sandy clay loam with a medium organic carbon content of 0.57 % a field capacity of 23.62 %, a permanent wilting point of 13.33 % and a bulk density of 1.33 mg m⁻³. The soil had an alkaline pH of 7.89 and an electrical conductivity of 0.213 d Sm⁻¹. The initial soil fertility status showed low available nitrogen (162 kg ha⁻¹), medium available phosphorus (16.4 kg ha⁻¹) and high available potassium (489 kg ha⁻¹).

Characterization of farm yard manure and poultry manure

The pH of FYM is slightly alkaline at 7.66, whereas PM has a more acidic nature with a pH of 6.46. This suggests that FYM may be more suitable for neutralizing acidic soils, while PM could be beneficial for balancing slightly alkaline soils. The electrical conductivity (EC), which indicates the salinity of the manure, is higher in PM (2.39 dSm⁻¹) compared to FYM (1.89 dSm⁻¹), reflecting a higher

concentration of soluble salts in PM. In terms of nutrient content, PM shows a significantly higher percentage of total nitrogen (2.30 %), total phosphorus (0.81 %) and total potassium (0.89 %), compared to FYM which contains 0.71 % nitrogen, 0.39 % phosphorus and 0.47 % potassium. These values suggest that PM could provide a more nutrient-dense fertilizer source, particularly for nitrogen-hungry crops. However, FYM has a slightly higher total organic carbon (TOC) content at 34.1 %, compared to 31.9 % in PM, indicating a higher organic matter content, which can improve soil structure and water retention over time.

Biologically, PM shows higher bacterial populations at 167×10^8 CFU g^{-1} , while FYM has 191×10^6 CFU g^{-1} . On the other hand, fungal populations are greater in FYM (89×10^4 CFU g^{-1}) compared to PM (106×10^3 CFU g^{-1}). Actinomycetes populations are notably higher in PM at 112×10^5 CFU g^{-1} compared to 64×10^5 CFU g^{-1} in FYM. These differences in microbial populations highlight the distinct microbiological environments promoted by each manure type, with PM favouring a richer bacterial and actinomycete community, while FYM supports a relatively higher fungal population.

Soil fertility parameters

The soil parameters, including pH, EC, available nitrogen (N), phosphorus (P) and potassium (K), were analyzed and documented in Table 1. All these parameters, except for EC, showed significant variation depending on the different sources of nutrient application. The highest pH value was observed in the absolute control treatment (8.03), while the lowest pH was recorded in the treatment where poultry manure at 75 % N equivalent, combined with Azophos and *Arbuscular mycorrhiza* (T_{10}), was applied (7.24). The soil EC did not show significant variations. However, the highest EC was recorded in the plot treated with poultry manure at 75 % N equivalent, combined with

Azophos and *Arbuscular mycorrhiza* (T_{10}) at 0.27 dS m^{-1} , while the absolute control (T_{11}) had the lowest EC at 0.12 dS m^{-1} .

The available nitrogen (N), phosphorus (P) and potassium (K) varied significantly with different nutrient applications. The highest available N (217 kg ha^{-1}) and P (26.1 kg ha^{-1}) were observed in the T_{10} treatment, which included poultry manure at 75 % N equivalent along with Azophos and *Arbuscular mycorrhiza*. In contrast, the absolute control (T_{11}) recorded the lowest values for N (169 kg ha^{-1}) and P (18.1 kg ha^{-1}). The highest available potassium (K) was measured in the T_7 treatment, which received FYM at 100 % N equivalent (534 kg ha^{-1}), while the lowest was recorded in the absolute control (T_{11}) at 488 kg ha^{-1} .

Soil enzyme activity

A comprehensive analysis data of the soil enzyme activity in the experimental field is given in Table 2. Soil microorganisms create the enzyme amylase, which converts starch into simpler carbohydrates. This process facilitates the breakdown of organic matter in the soil and contributes to the cycling of carbon by increasing the accessibility of complex organic compounds to microbes. The soil amylase activity varied between 0.81 and $1.21 \text{ mg reducing sugars } g^{-1} \text{ soil } 24 \text{ h}^{-1}$. The T_{10} treatment exhibited the highest amylase activity at $1.21 \text{ mg reducing sugars } g^{-1} \text{ soil } 24 \text{ h}^{-1}$, whereas the lowest activity, at $0.81 \text{ mg reducing sugars } g^{-1} \text{ soil } 24 \text{ h}^{-1}$, was recorded in the T_{11} treatment.

An important measure of microbial activity and their capacity to hydrolyze sucrose into glucose and fructose is the activity of the invertase enzyme in soil. Numerous soil microorganisms, such as fungus and bacteria, generate this enzyme. An understanding of the diversity and functional abilities of the soil microbial population can be gained by tracking invertase activity. At

Table 1. Impact of inorganics, organics and bio fertilizers on soil properties of fodder sorghum cultivated field.

Treatment Details	pH	EC (dSm ⁻¹)	Available N (kg ha ⁻¹)	Available P (kg ha ⁻¹)	Available K (kg ha ⁻¹)
T ₁ - RDF	7.56 ± 0.1	0.25 ± 0.03	175 ± 0.9	19.5 ± 0.4	507 ± 8.8
T ₂ - Soil test value based recommendation	7.71 ± 0.7	0.23 ± 0.06	172 ± 2.5	18.2 ± 0.7	492 ± 3.8
T ₃ - <i>Arbuscular mycorrhiza</i> + T ₂	7.45 ± 0.2	0.13 ± 0.01	186 ± 0.7	22.2 ± 0.2	497 ± 5.9
T ₄ - Azophos + T ₂	7.9 ± 0.01	0.25 ± 0.03	184 ± 3.5	23.5 ± 0.5	491 ± 2.9
T ₅ - T ₂ + (Azophos + <i>Arbuscular mycorrhiza</i>)	7.64 ± 0.3	0.21 ± 0.06	193 ± 3.2	25.2 ± 0.3	512 ± 8.1
T ₆ - RDF at 75 % N + (Azophos + <i>Arbuscular mycorrhiza</i>)	7.49 ± 0.2	0.26 ± 0.01	202 ± 1.9	24.3 ± 0.1	504 ± 1.1
T ₇ - FYM N equivalent basis (100 % N)	7.55 ± 0.1	0.15 ± 0.01	194 ± 0.2	23.5 ± 0.8	534 ± 9.1
T ₈ - PM N equivalent basis (100 % N)	7.49 ± 0.3	0.22 ± 0.01	199 ± 1.2	24.6 ± 0.2	530 ± 2.7
T ₉ - FYM N equivalent basis (75 % N) + (Azophos + <i>Arbuscular mycorrhiza</i>)	7.45 ± 0.1	0.16 ± 0.06	204 ± 1.4	23.9 ± 0.5	495 ± 6.4
T ₁₀ - PM N equivalent basis (75 % N) + (Azophos + <i>Arbuscular mycorrhiza</i>)	7.42 ± 0.4	0.27 ± 0.03	217 ± 4.1	26.1 ± 0.9	523 ± 7.3
T ₁₁ - Absolute Control	8.03 ± 0.2	0.12 ± 0.09	169 ± 2.5	18.1 ± 0.3	488 ± 3.4
Mean	7.59 ± 0.3	0.22 ± 0.02	190 ± 0.5	22.58 ± 0.8	508 ± 2.6
SE(d)	0.14	0.07	4.15	0.41	11.5
CD (P=0.05)	0.3	NS	8.72	0.86	24.2

Table 2. Impact of inorganics, organics and bio fertilizers on soil enzyme activities of fodder sorghum cultivated field.

Treatment Details	Amylase mg reducing sugars g ⁻¹ soil 24 h ⁻¹	Invertase mg reducing sugars g ⁻¹ soil 24 h ⁻¹	Cellulase µg reducing sugars g ⁻¹ soil 24 h ⁻¹	Phosphatase µg phenols g ⁻¹ soil 24 h ⁻¹	Dehydrogenase µg triphenyl formazan g ⁻¹ soil 24 h ⁻¹	Urease (mg NH ₄ - N g ⁻¹ soil 24 h ⁻¹)
Initial Soil Sample	0.78 ± 0.02	0.66 ± 0.01	15.3 ± 0.1	11.63 ± 0.14	6.72 ± 0.12	5.71 ± 0.23
T ₁ - RDF	0.85 ± 0.02	0.76 ± 0.02	18.5 ± 0.3	15.14 ± 0.25	8.53 ± 0.05	6.51 ± 0.06
T ₂ - Soil test value based recommendation	0.95 ± 0.02	0.82 ± 0.01	22.1 ± 0.2	16.92 ± 0.34	7.74 ± 0.05	6.43 ± 0.09
T ₃ - <i>Arbuscular mycorrhiza</i> + T ₂	0.88 ± 0.02	0.85 ± 0.01	23.2 ± 0.4	15.28 ± 0.28	9.18 ± 0.06	11.12 ± 0.08
T ₄ - Azophos + T ₂	0.96 ± 0.02	0.95 ± 0.02	25.8 ± 0.5	15.15 ± 0.22	9.52 ± 0.08	13.23 ± 0.26
T ₅ - T ₂ + (<i>Azophos</i> + <i>Arbuscular mycorrhiza</i>)	0.89 ± 0.02	0.94 ± 0.02	27.6 ± 0.3	15.83 ± 0.06	10.11 ± 0.09	14.67 ± 0.06
T ₆ - RDF at 75 % N + (<i>Azophos</i> + <i>Arbuscular mycorrhiza</i>)	0.86 ± 0.02	1.27 ± 0.01	32.3 ± 0.6	18.9 ± 0.16	9.19 ± 0.05	15.24 ± 0.06
T ₇ - FYM N equivalent basis (100 % N)	0.99 ± 0.01	1.13 ± 0.03	28.6 ± 0.2	17.01 ± 0.01	8.71 ± 0.14	15.91 ± 0.32
T ₈ - PM N equivalent basis (100 % N)	0.99 ± 0.04	1.35 ± 0.03	33.1 ± 0.3	19.11 ± 0.08	11.01 ± 0.07	16.22 ± 0.16
T ₉ - FYM N equivalent basis (75 % N) + (<i>Azophos</i> + <i>Arbuscular mycorrhiza</i>)	0.97 ± 0.02	1.07 ± 0.02	27.7 ± 0.2	19.56 ± 0.09	10.86 ± 0.09	18.21 ± 0.2
T ₁₀ - PM N equivalent basis (75 % N) + (<i>Azophos</i> + <i>Arbuscular mycorrhiza</i>)	1.12 ± 0.01	1.45 ± 0.02	34.8 ± 0.5	19.73 ± 0.06	11.56 ± 0.21	17.92 ± 0.3
T ₁₁ - Absolute Control	0.81 ± 0.01	0.77 ± 0.02	18.2 ± 0.3	13.31 ± 0.1	8.04 ± 0.06	6.42 ± 0.13
Mean	0.93 ± 0.01	1.03 ± 0.01	26.4 ± 0.4	16.9 ± 0.3	9.45 ± 0.04	12.72 ± 0.09
SE(d)	0.03	0.024	0.626	0.317	0.237	0.28
CD (P=0.05)	0.062	0.05	1.315	0.666	0.499	0.588

the end of the fifth harvest of green fodder, the average soil invertase activity varied between 0.77 and 1.45 mg reducing sugars g⁻¹ soil 24 h⁻¹. T₁₀ had the maximum invertase activity (1.45 mg reducing sugars g⁻¹ soil 24 h⁻¹), whereas T₁₁ had the lowest activity (0.77 mg reducing sugars g⁻¹ soil 24 h⁻¹).

Cellulase activity serves as a critical indicator for evaluating soil health and the overall biological activity of soil. A robust soil ecosystem with active cellulase-producing microorganisms tends to exhibit greater resilience to environmental stresses and disturbances. Cellulase activity in soil varied from 18.2 to 34.8 µg of reducing sugars g⁻¹ soil 24 h⁻¹. T₁₁ had the lowest activity (18.2 µg reducing sugars g⁻¹ soil 24 h⁻¹ and T₁₀ had the highest activity (34.8 µg reducing sugars g⁻¹ soil 24 h⁻¹).

Phosphatase enzymes, both acid and alkaline, play a vital role in phosphorus mineralization, making it accessible for plant uptake. The mean soil phosphatase activity ranged from 13.31 to 19.73 µg phenols g⁻¹ soil 24 h⁻¹. After the 5th green fodder harvest, the highest phosphatase activity was observed in T₁₀ (19.73 µg phenols g⁻¹ soil 24 h⁻¹), while the lowest activity was recorded in the absolute control T₁₁ (13.31 µg phenols g⁻¹ soil 24 h⁻¹).

Dehydrogenase activity serves as a key indicator of overall microbial activity and soil health, crucial for organic matter decomposition. The soil dehydrogenase activity ranged from 7.74 to 11.56 µg triphenyl formazan g⁻¹ soil 24 h⁻¹. Among the treatments, the highest activity was recorded in T₁₀ (11.56 µg triphenyl formazan g⁻¹ soil 24 h⁻¹), while the lowest was observed in T₂ soil test value based

recommended dose received treatment (7.74 µg triphenyl formazan g⁻¹ soil 24 h⁻¹). Elevated nutrient levels, particularly nitrogen (N), enhanced dehydrogenase activity, indicating increased oxidative activity among soil microflora and microbial populations (26).

Nitrogen cycling depends on urea's breakdown into ammonia, which is catalysed by urease. The average amount of urease in the soil varied between 17.92 and 6.42 mg NH₄- N g⁻¹ soil 24 h⁻¹. At the end of 5th fodder harvesting, T₁₀ had the maximum soil urease activity (17.92 mg NH₄- N g⁻¹ soil 24 h⁻¹), whereas T₁₁ absolute control had the lowest value (6.42 mg NH₄- N g⁻¹ soil 24 h⁻¹).

Soil microbial population

During the final fodder harvest, the mean bacterial population (Fig. 1) varied across treatments, ranging from 56.2 to 39.5 × 10⁶ CFU g⁻¹ of soil. The soil treated with poultry manure at 75 % N equivalent, supplemented with *Azophos* and *Arbuscular mycorrhiza*, exhibited the highest bacterial population (56.2 × 10⁶ CFU g⁻¹ of soil) compared to other treatments. In contrast, the lowest bacterial population was observed in the absolute control treatment, which recorded 39.5 × 10⁶ CFU g⁻¹ of soil.

The fungal communities in the experimental field are shown in detail in Fig. 2, with mean populations ranging from 31.5 to 14.7 × 10⁴ CFU g⁻¹ of soil across treatments. Treatment T₁₀ exhibited the highest fungal population (31.5 × 10⁴ CFU g⁻¹), while the lowest (14.7 × 10⁴ CFU g⁻¹) was found in the T₁₁ absolute control at the end of final fodder harvest. Fig. 3 illustrates the actinomycetes populations in the experimental field, ranging from 20.1 to 8.9 × 10² CFU g⁻¹ of soil at the end of final fodder sorghum

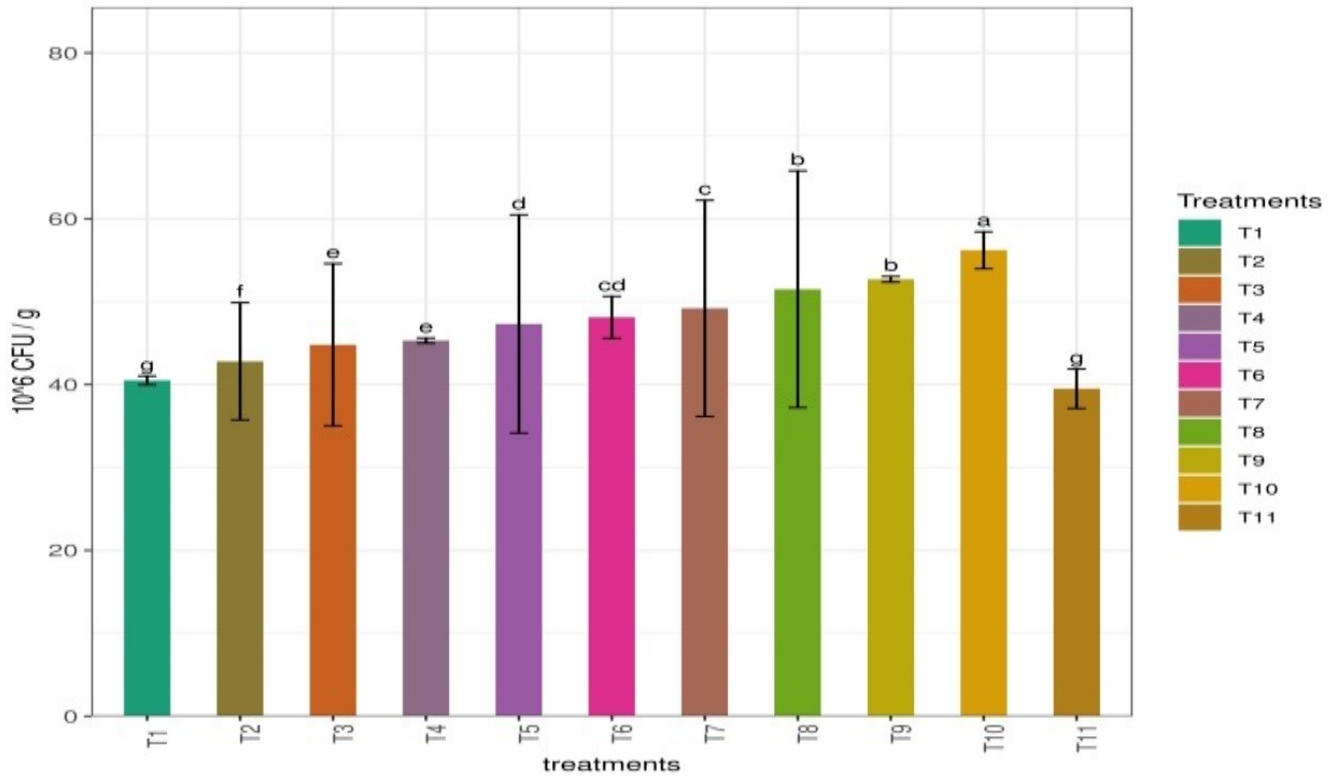


Fig. 1. Impact of inorganics, organics and bio fertilizers on soil bacterial population (10⁶ CFU g⁻¹) of fodder sorghum cultivated field.

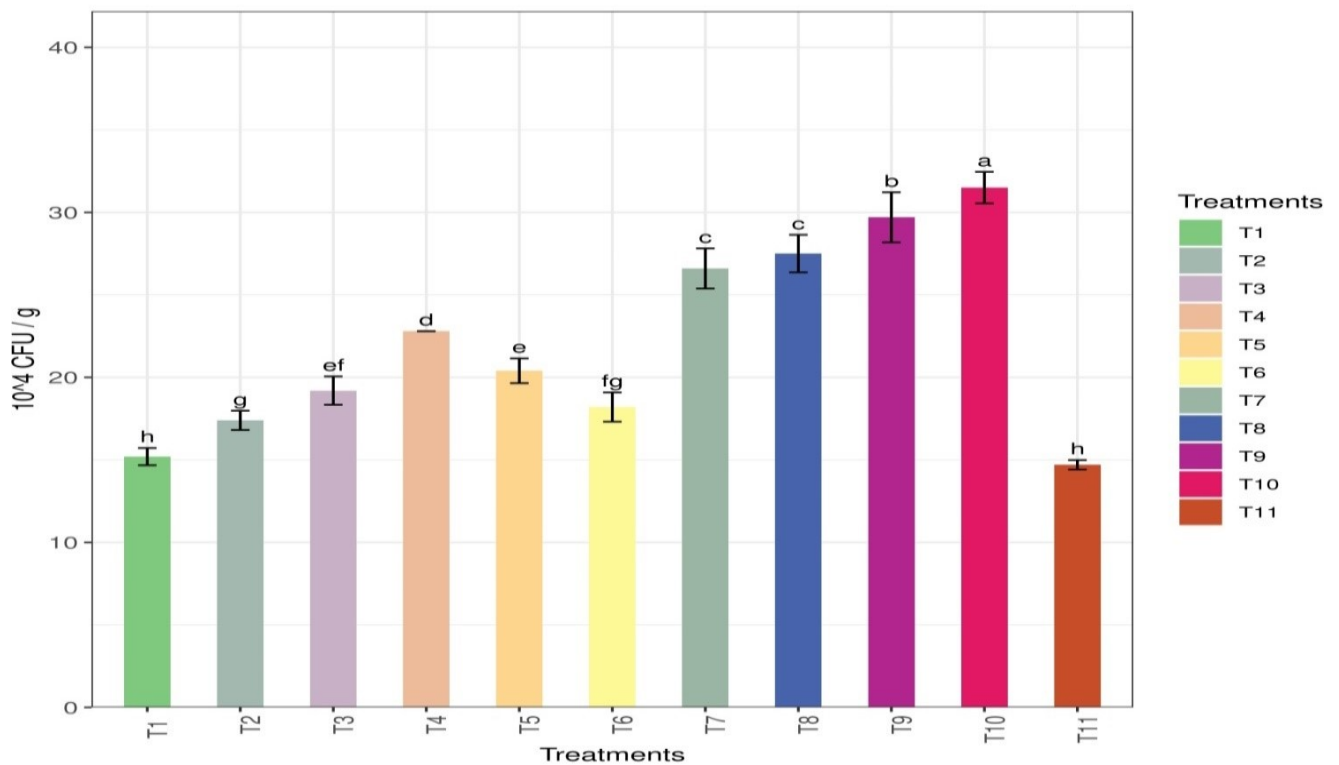


Fig. 2. Impact of inorganics, organics and bio fertilizers on soil fungal population (10⁴ CFU g⁻¹) of fodder sorghum cultivated field.

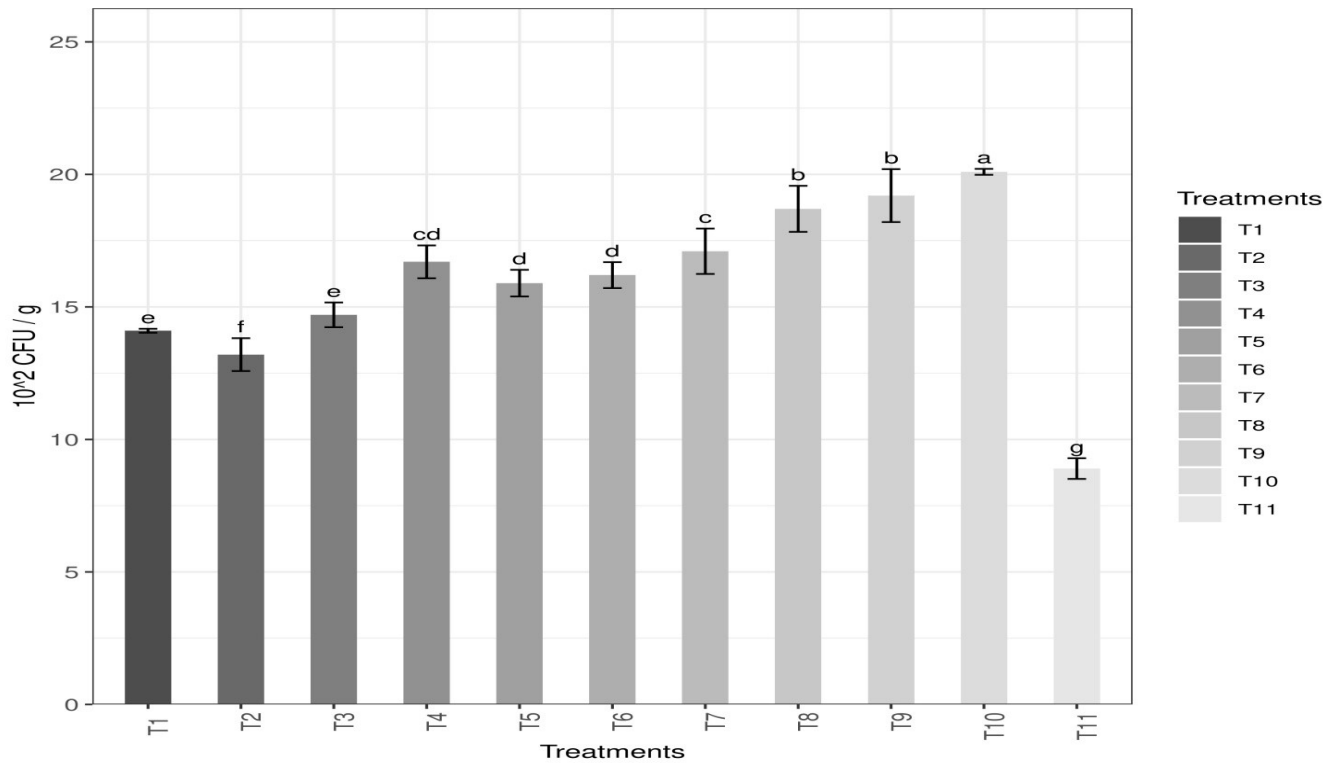


Fig. 3. Impact of inorganics, organics and bio fertilizers on soil actinomycetes population (10^2 CFU g^{-1}) of fodder sorghum cultivated field.

harvest. Poultry manure (PM) treatments generally showed higher actinomycetes populations compared to inorganic fertilizers and farmyard manure (FYM). Treatment T₁₀ had the highest actinomycetes population (20.1×10^2 CFU g^{-1} soil), while the lowest (8.9×10^2 CFU g^{-1} soil) was recorded in the T₁₁ absolute control plot at the final harvesting stage.

Soil carbon

Detailed overview of Soil Organic Carbon (SOC) content is given in Fig. 4. SOC serves as a crucial energy source for

microorganisms within terrestrial ecosystems, playing a pivotal role in ecosystem dynamics by influencing soil structure and enhancing productivity. Soil organic carbon (SOC) levels at the end of the seventh fodder harvest ranged from 0.51 - 0.70 % depending on the treatment. The treatment with the greatest SOC content was recorded in T₁₀ applied 75 % N equivalent of poultry manure (PM) coupled with *Azaphos* and *Arbuscular mycorrhiza* and its SOC content was 0.70 %. T₉, which used farmyard manure (FYM) at 75 % N equivalent with *Azaphos* and *Arbuscular mycorrhiza* and had a SOC level of 0.68 %, trailed closely

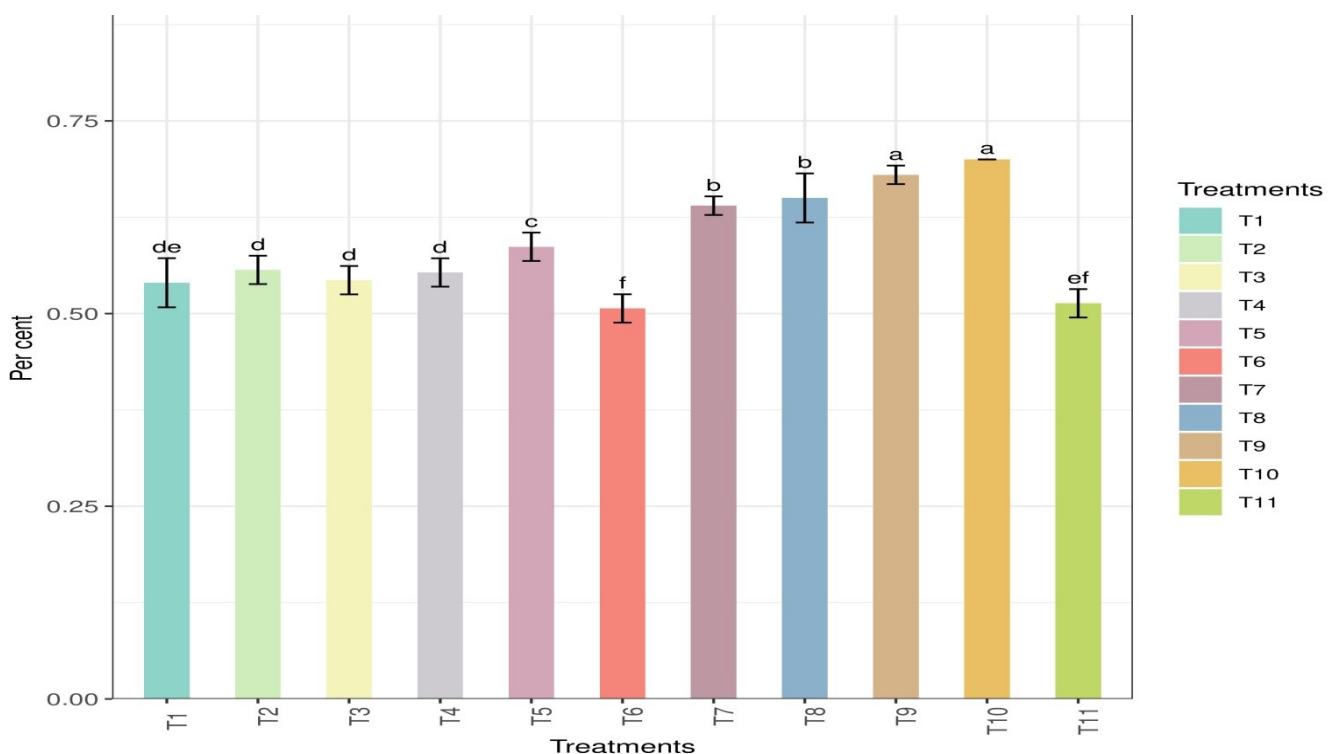


Fig. 4. Impact of inorganics, organics and bio fertilizers on soil organic carbon (%) in fodder sorghum cultivated field.

Table 3. Impact of inorganics, organics and bio fertilizers on soil carbon fractions and balance of fodder sorghum cultivated field.

Treatment Details	Walkley and Black C (mg kg ⁻¹) (1)	Water-soluble carbon (mg kg ⁻¹) (2)	KMnO ₄ carbon (Active C) (mg kg ⁻¹) (3)	Bulk density (Mg m ⁻³) (4)	Passive carbon (mg/kg) (5) = (1) - (2+3)	PSOC (%) (6)	Permanent soil carbon stock (PSCS) (t ha ⁻¹ year ⁻¹) (7) = (6)*(4)* Depth	Added SOC in passive carbon pool (t ha ⁻¹ year ⁻¹) (8) = Initial PSCS - treatment PSCS
Initial Soil Sample	5200 ± 104	875 ± 9.67	865 ± 9.3	1.33 ± 0.03	3460 ± 63.8	0.35 ± 0.01	6.91 ± 0.13	
T ₁ - RDF	5400 ± 45	1120 ± 0.96	980 ± 15.4	1.33 ± 0.02	3300 ± 30.9	0.33 ± 0.01	6.59 ± 0.11	-0.32 ± 0.01
T ₂ - Soil test value based recommendation	5530 ± 79	870 ± 5.92	665 ± 12.4	1.31 ± 0.03	3995 ± 57.7	0.39 ± 0.01	7.86 ± 0.02	0.96 ± 0.01
T ₃ - AM + T ₂	5430 ± 18	730 ± 4.35	695 ± 0.3	1.33 ± 0.03	4005 ± 30.6	0.40 ± 0.01	7.99 ± 0.08	1.09 ± 0.02
T ₄ - Azophos + T ₂	5503 ± 97	720 ± 14.8	765 ± 15.9	1.32 ± 0.01	4018 ± 25.6	0.41 ± 0.01	7.96 ± 0.02	1.06 ± 0.02
T ₅ - T ₂ + (Azophos + AM)	5933 ± 107	752 ± 13.8	830 ± 7.7	1.31 ± 0.02	4287 ± 22.4	0.42 ± 0.01	8.32 ± 0.14	1.67 ± 0.02
T ₆ - RDF at 75 % N + (Azophos + AM)	5735 ± 124	608 ± 6.9	614 ± 1.8	1.34 ± 0.03	4453 ± 22.9	0.45 ± 0.01	9.07 ± 0.15	1.82 ± 0.01
T ₇ - FYM N equivalent basis (100 % N)	6380 ± 33	821 ± 4.5	881 ± 9.2	1.32 ± 0.02	4699 ± 42.2	0.47 ± 0.01	9.49 ± 0.02	2.65 ± 0.03
T ₈ - PM N equivalent basis (100 % N)	6656 ± 66	756 ± 10.9	934 ± 1.6	1.37 ± 0.01	4785 ± 84.2	0.48 ± 0.01	9.88 ± 0.04	2.92 ± 0.04
T ₉ - FYM N equivalent basis (75 % N) + (Azophos + AM)	6825 ± 32	1326 ± 15.4	950 ± 2.1	1.38 ± 0.01	4446 ± 81.3	0.45 ± 0.01	9.26 ± 0.19	2.38 ± 0.05
T ₁₀ - PM N equivalent basis (75 % N) + (Azophos + AM)	7057 ± 5.9	937 ± 18.3	1127 ± 13.3	1.37 ± 0.01	4896 ± 43.5	0.49 ± 0.01	10.23 ± 0.16	3.06 ± 0.06
T ₁₁ - Absolute Control	4947 ± 65	834 ± 13.8	958 ± 11.1	1.29 ± 0.03	3314 ± 18.4	0.35 ± 0.01	6.58 ± 0.11	-0.27 ± 0.01
Mean	6014 ± 9.3	859 ± 1.75	842 ± 2.87	1.34 ± 0.01	4332 ± 19.9	0.44 ± 0.01	8.69 ± 0.04	1.73 ± 0.01
SE(d)	110.914	19.66	16.67	0.03	84.723	0.008	0.197	0.048
CD (P=0.05)	232.992	41.298	35.014	NS	177.974	0.016	0.407	0.101

behind. In contrast, the lowest SOC level of 0.51 % was observed in T₁₁, the absolute control treatment.

Soil carbon fractions

The findings regarding soil carbon fraction are summarized in Table 3, highlighting notable differences among treatments. The highest level of water-soluble carbon was observed in T₉, where farmyard manure was applied at 75 % N equivalent, supplemented with *Arbuscular mycorrhiza* and Azophos (1326 mg kg⁻¹). In contrast, the lowest concentration was found in T₁₁, the absolute control treatment (720 mg kg⁻¹). Similarly, notable variations were observed in Walkley and Black carbon and KMnO₄ oxidizable carbon (active pool carbon). Treatment T₁₀, which involved poultry manure (PM) at 75 % N equivalent with Azophos and *Arbuscular mycorrhiza*, displayed the highest soil SOC at 6957 mg kg⁻¹. In contrast, treatment T₄, based on soil test recommendations and supplemented with Azophos, exhibited the lowest SOC level at 4947 mg kg⁻¹.

Treatment T₁₀, which involved poultry manure (PM) at 75 % N equivalent with Azophos and *Arbuscular mycorrhiza* exhibited the highest KMnO₄ oxidizable carbon at 1127 mg kg⁻¹, whereas the lowest was recorded in T₆, which applied recommended doses of fertilizer (RDF) at 75 % N with *Arbuscular mycorrhiza* and Azophos, registering 614 mg kg⁻¹. The treatment with the highest soil passive carbon content, measured in T₁₀ (4896 mg kg⁻¹), the lowest passive carbon content was observed in T₁, which received the recommended dose of fertilizer (3314 mg kg⁻¹)

Carbon balance

The existence of a stable and persistent stock of passive soil carbon is essential for maintaining long-term environmental health, promoting agricultural sustainability and facilitating effective climate regulation. This enduring carbon reservoir plays a significant role in mitigating climate change, enhancing soil health and fertility, supporting biodiversity and enabling sustainable land management practices. The average permanent soil carbon stock ranged from 6.58 to 10.23 t ha⁻¹ year⁻¹ across treatments. The highest permanent soil carbon stock, at 10.23 t ha⁻¹ year⁻¹, was observed in T₁₀, which involved poultry manure (PM) at 75 % N equivalent with Azophos and *Arbuscular mycorrhiza*. The lowest value of 6.58 t ha⁻¹ year⁻¹ was noted in T₁, where the recommended dose of fertilizer was applied. The average increase in soil organic carbon in the passive pool varied from -0.32 to 3.06 t ha⁻¹ year⁻¹. Treatment T₁₀, involving poultry manure (PM) at 75 % N equivalent with Azophos and *Arbuscular mycorrhiza*, recorded the highest value of 3.06 t ha⁻¹ year⁻¹, while, treatment T₁₁, the absolute control, had the lowest value at -0.32 t ha⁻¹ year⁻¹.

Soil Carbon Stock

Preserving and enhancing carbon stocks are essential for mitigating atmospheric CO₂ levels, enhancing soil fertility and bolstering ecosystem resilience. Among the various treatments, T₁₀ sequestered 14.11 t ha⁻¹ year⁻¹ of carbon, followed closely by T₉ with 13.97 t ha⁻¹ year⁻¹, showcasing the highest soil carbon stocks (Fig. 5). In contrast, the lowest carbon stock of 10.17 t ha⁻¹ year⁻¹ was observed in

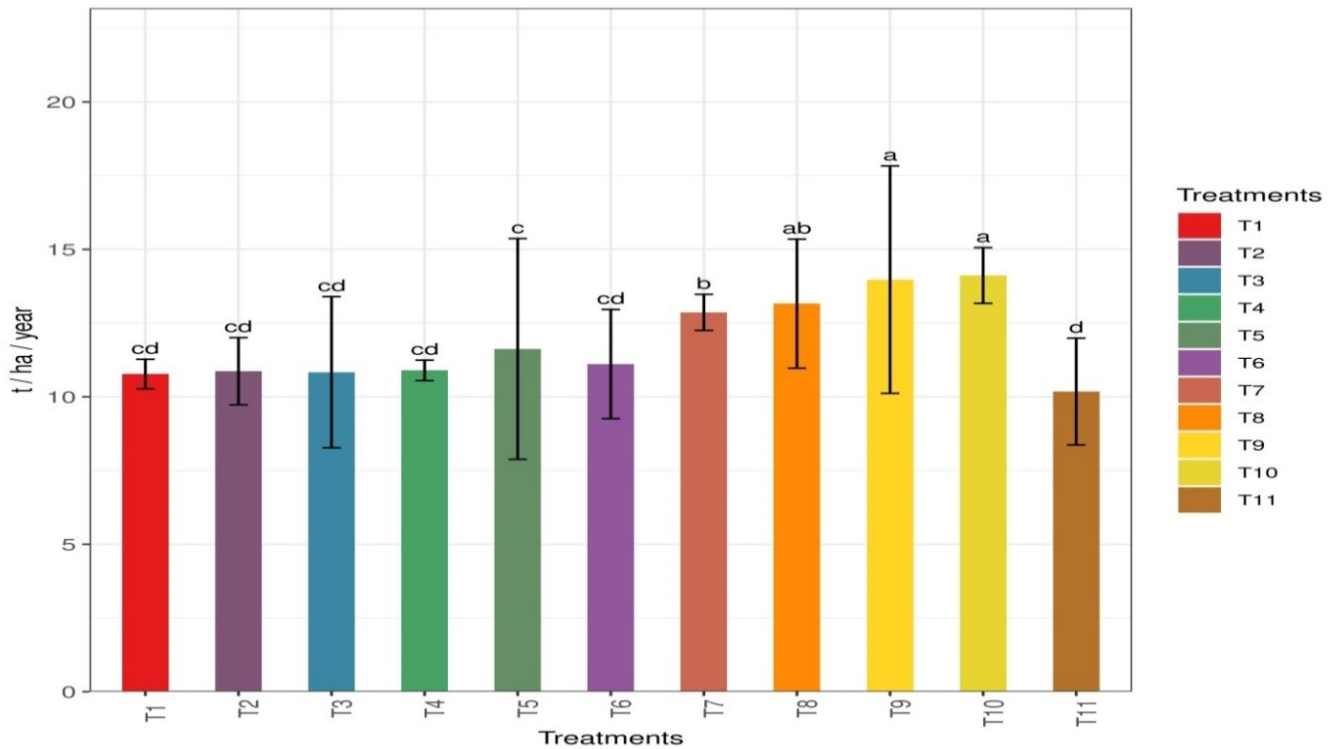


Fig. 5. Impact of inorganics, organics and bio fertilizers on Carbon stock ($t\ ha^{-1}\ year^{-1}$) of fodder sorghum cultivated field.

the absolute control plot, T₁₁. The higher carbon stocks in treatments like T₁₀ are likely due to practices involving biomass addition and the storage of recalcitrant carbon. Carbon is often associated with microaggregates, which shield it from degradation and facilitate long-term storage (27). This protective mechanism likely contributed to the elevated carbon stock observed in T₁₀.

Yield and quality parameters

The fodder yield of multi cut sorghum was significantly higher when nutrients were applied to the fodder crop compared to the control plot (Table 4). Particularly, applying poultry manure at a 75 % nitrogen equivalent

basis + (Azophos + *Arbuscular mycorrhiza*) and farm yard manure at a 75 % nitrogen equivalent basis + (Azophos + *Arbuscular mycorrhiza*) significantly increased the fodder yield of multi cut sorghum (164 and 156 $t\ ha^{-1}$) over the absolute control from (125 $t\ ha^{-1}$).

Nutrient application to the crop significantly increased dry matter production compared to the control plot. Specifically, the application of poultry manure at 75 % nitrogen equivalent combined with Azophos and *Arbuscular mycorrhiza* resulted in a substantial increase in dry matter production (39.9 $t\ ha^{-1}$) compared to the absolute control (29.2 $t\ ha^{-1}$). This was followed by farmyard manure at 75 % nitrogen equivalent with

Table 4. Impact of inorganics, organics and bio fertilizers on yield and quality of fodder sorghum.

Treatment Details	Green fodder Yield ($t\ ha^{-1}\ year^{-1}$)	Dry matter ($t\ ha^{-1}\ year^{-1}$)	Protein content (%)	Crude fiber (%)
T ₁ - RDF	147 ± 3.1	34.5 ± 0.3	7.16 ± 0.1	23.1 ± 0.31
T ₂ - Soil test value based recommendation	137 ± 2.3	32.3 ± 0.4	7.24 ± 0.2	23.2 ± 0.26
T ₃ - Arbuscular mycorrhiza + T ₂	140 ± 0.2	33.2 ± 0.1	7.23 ± 0.1	21.7 ± 0.38
T ₄ - Azophos + T ₂	139 ± 1.5	32.8 ± 0.3	7.42 ± 0.1	20.1 ± 0.26
T ₅ - T ₂ + (Azophos + Arbuscular mycorrhiza)	147 ± 2.3	34.7 ± 0.7	7.97 ± 0.1	21.5 ± 0.19
T ₆ - RDF at 75 % N + (Azophos + Arbuscular mycorrhiza)	148 ± 1.3	33.6 ± 0.6	7.83 ± 0.1	23.4 ± 0.49
T ₇ - FYM N equivalent basis (100 % N)	150 ± 0.6	34.7 ± 0.5	7.98 ± 0.1	25.5 ± 0.36
T ₈ - PM N equivalent basis (100 % N)	155 ± 2.9	34.8 ± 0.6	8.02 ± 0.1	23.2 ± 0.32
T ₉ - FYM N equivalent basis (75 % N) + (Azophos + Arbuscular mycorrhiza)	156 ± 2.3	35.9 ± 0.7	8.19 ± 0.1	24.8 ± 0.34
T ₁₀ - PM N equivalent basis (75 % N) + (Azophos + Arbuscular mycorrhiza)	164 ± 1.5	39.9 ± 0.7	8.56 ± 0.1	21.9 ± 0.34
T ₁₁ - Absolute Control	125 ± 1.1	29.2 ± 0.3	6.12 ± 0.4	20.6 ± 0.23
Mean	146 ± 0.4	34.3 ± 0.2	7.61 ± 0.5	22.7 ± 0.02
SE(d)	3.47	0.86	0.16	0.58
CD (P=0.05)	7.29	1.8	0.33	1.22

Azophos and *Arbuscular mycorrhiza* (35.9 t ha^{-1}) and poultry manure at 100 % nitrogen equivalent (34.8 t ha^{-1}) which were statistically similar to each other (Table 4). Application of poultry manure at N equivalent basis (75 % N) along with Azophos and *Arbuscular mycorrhiza* (T_{10}) applied treatment significantly increased (8.56 %) protein content over absolute control from (6.12 %). When FYM was applied at 100 % N equivalent basis, the crude fibre content was much greater (25.6 %) than in the control plot (20.6 %).

Discussion

The integration of poultry manure and biofertilizers, including *Arbuscular Mycorrhiza* (AM) fungi and Azophos, has gained attention for its positive effects on soil health, nutrient availability and plant growth in the present investigation. This approach not only improves essential soil properties, but also significantly enhances the microbial ecosystem, fostering a dynamic and nutrient-rich environment. Poultry manure serves as a rich source of organic nutrients, while biofertilizers boost microbial activity and nutrient cycling. Together, they create a synergistic effect that supports sustainable farming practices, improves soil fertility and contributes to long-term carbon sequestration.

The application of poultry manure and biofertilizers resulted in slightly decrease the soil pH. Poultry manure, can slightly acidify the soil, while the biofertilizers (AM fungi and Azophos) may buffer this effect, maintaining soil pH by improving nutrient cycling (28). The combination of these inputs also increased the electrical conductivity (EC) of the soil with less percentage. This is likely due to the addition of salts and nutrients from poultry manure, which increases the soil's ionic content (29). The availability of essential nutrients such as nitrogen (N), phosphorus (P) and potassium (K) was greatly improved by the integrated use of poultry manure and biofertilizers, increasing by 28.40 %, 44.19 % and 9.43 % respectively. Poultry manure serves as a rich source of organic N, P and K and its decomposition releases these nutrients into forms that plants can absorb. Azophos, which contains *Azospirillum* and phosphate-solubilizing bacteria (PSB), enhances nitrogen fixation and solubilizes phosphorus, making them more readily available to plants. AM fungi improve nutrient uptake by increasing root surface area and enhancing symbiotic nutrient exchanges, thus promoting higher nutrient availability (30).

The use of poultry manure, *Arbuscular mycorrhiza* (AM) fungi and Azophos has been shown to significantly boost soil enzymes activity viz., amylase, invertase, cellulase phosphatase, dehydrogenase and urease, a synergistic effect that improves overall soil health. Since, poultry manure provides a rich source of organic matter, which enhances microbial activity, leading to an increase in microbial biomass and diversity. This organic nitrogen supply also stimulates urease activity, a key enzyme in nitrogen cycling, while the overall increase in microbial activity promotes the production of several important soil enzymes, fostering a more fertile and dynamic

environment (31).

AM fungi play a vital role by forming beneficial relationships with plant roots, enhancing root exudation and encouraging positive microbial interactions in the rhizosphere. These interactions help promote the activity of invertase, an enzyme crucial for carbon cycling, by breaking down sucrose into simpler sugars. Additionally, AM fungi and Azophos enhance phosphorus uptake and stimulate the production of phosphatase, further increasing phosphorus availability in the soil (32). Azophos, a biofertilizer containing *Azospirillum* and phosphate-solubilizing bacteria, complements these benefits by promoting higher microbial activity and diversity. It works synergistically with phosphatase to increase phosphorus availability and supports nitrogen fixation, boosting nitrogen levels in the soil, which, in turn, enhances urease activity (33).

The increased microbial populations resulting from this combination also elevate amylase activity, which is influenced by factors such as soil pH, nutrient levels and microbial biomass. The observed increase in amylase activity in treatment T_{10} is a direct result of the rise in microbial populations due to the combined use of these soil amendments (34). This approach not only improves soil fertility but also supports sustainable farming by promoting efficient nutrient cycling.

The microbial community, including bacteria, fungi and actinomycetes, showed significant improvements in diversity and activity due to the application of poultry manure and biofertilizers. Poultry manure, rich in organic matter, provided a conducive environment for microbial growth, while AM fungi improved the mycorrhizal network, enhancing nutrient uptake and supporting the microbial population (35, 36). This enriched microbial diversity promotes better nutrient cycling and soil fertility, key to sustainable fodder sorghum production.

Soil organic carbon (SOC) was boosted by 39.22 % with the combined application of poultry manure and biofertilizers. The increased SOC was attributed to improved microbial activity and the addition of organic matter from poultry manure, which enhanced soil temperature, moisture and humus content (37, 38). Permanent soil carbon stock also increased by 38.74 %, contributing to long-term carbon sequestration (39). The decomposition of poultry manure and the role of AM fungi in root biomass formation stabilized organic matter, promoting soil aggregation and boosting carbon storage (40, 41).

Green fodder yield, DMP, protein content and crude fiber content were significantly enhanced by 31.2 %, 36.4 % and 39.9 % respectively, with the application of poultry manure, AM fungi and Azophos. This synergistic effect is due to improved nutrient availability, nitrogen fixation and phosphate uptake. The protein content was notably increased due to the nitrogen fixation by Azophos, further supplemented by organic nitrogen from poultry manure. AM fungi facilitated better nutrient uptake, ensuring optimal plant growth and biomass production (42). A

study suggests that integrating poultry manure with biofertilizers such as AM fungi and Azophos creates a highly efficient nutrient management system that enhances soil health, promotes microbial diversity, boosts nutrient availability and improves the yield and quality of fodder sorghum (43). This sustainable approach aligns with carbon sequestration goals, contributing to long-term environmental benefits and sustainable agricultural practices.

Conclusion

According to the study's findings, the combined application of poultry manure at 75 % N equivalent basis along with *Arbuscular mycorrhiza* (AM) fungi and Azophos significantly enhances the yield, quality and soil health in multicut fodder sorghum. This integrated nutrient management strategy boosts green fodder yield, DMP, protein content and crude fiber content by leveraging the synergistic effects of organic nutrients, enhanced nitrogen fixation and improved nutrient uptake. The soil parameters such as electrical conductivity, pH and available N, P and K levels show marked differences, indicating better nutrient availability and soil fertility, enhanced soil enzyme activities, such as amylase, invertase and phosphatase, reflect increased microbial activity and nutrient cycling. The microbial community becomes more diverse and active, further contributing to soil health. Soil organic carbon (SOC) and soil carbon stock are significantly increased, promoting long-term carbon sequestration. This holistic approach not only improves crop productivity but also supports sustainable agricultural practices, ensuring long-term soil health and environmental benefits.

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Authors' contributions

NS carried out the experiment, took observations and analysed the data. KS guided the research by formulating the research concept, helped in securing research funds and approved the final manuscript. RJ contributed by developing the ideas, reviewed the manuscript and helped in procuring research grants. RK and KN contributed by imposing the experiment, helped in editing, summarizing and revising the manuscript. RK and RA helped in editing, summarizing and revising the manuscript.

Compliance with ethical standards

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

Ethical issues: None.

Declaration of generative AI and AI-assisted technologies in the writing process

During the preparation of this work the author(s) not used AI tools and the author(s) reviewed and edited the content as needed and take(s) full responsibility for the content of the publication.

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