



Impact of Sugarcane Residue-based Amendments on Soil Organic Carbon Stock and Yield of Black Gram (*Vigna mungo* L.) under Natural Farming Protocols

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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Abstract

Declining soil organic carbon and increasing reliance on synthetic fertilizers are important constraints to sustainable crop production. The present field experiment was conducted during the kharif season at Surat, Gujarat, India, to evaluate the influence of sugarcane residue-based amendments and natural farming practices on soil properties, nutrient uptake, soil organic carbon stock, and yield of black gram (*Vigna mungo* L.). The experiment was laid out in a randomized block design with six treatments and three replications. The treatments consisted of control, sugarcane trash mulching, sugarcane bagasse compost, press mud application, natural farming with biochar, and recommended dose of fertilizers. Soil samples were collected before sowing and after harvest for analysis of soil organic carbon, pH, electrical conductivity, available

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nitrogen, available phosphorus, available potassium, and bulk density. Plant samples were analyzed for nitrogen, phosphorus, and potassium uptake at harvest. The results showed that sugarcane residue-based amendments improved soil organic carbon, nutrient availability, bulk density, nutrient uptake, and crop yield compared with the untreated control. Among the treatments, natural farming with biochar recorded the highest soil organic carbon content (0.68%), grain yield (1328 kg ha⁻¹), and biomass yield (3025 kg ha⁻¹). The same treatment also recorded the highest final soil organic carbon stock (13.36 t C ha⁻¹) and carbon accumulation (4.04 t C ha⁻¹). Press mud application also showed improvement in soil organic carbon and carbon stock. The findings indicate that recycling sugarcane residues through organic amendments, particularly when combined with biochar-based natural farming inputs, can improve soil health and support black gram productivity under the tested conditions. However, longer-term evaluation is needed to confirm the persistence of these effects across seasons and soil types.

Keywords: Biochar amendment; soil organic carbon stock; soil health; sustainable agriculture; sugarcane residues; Vigna mungo.

Nomenclature & Abbreviations

Soil Chemical & Physical Metrics:

| | |
|----------------------------|---|
| <i>SOC</i> | : Soil Organic Carbon (%) |
| <i>SOC Stock</i> | : Soil Organic Carbon Stock (t C ha ⁻¹) |
| <i>Carbon Accumulation</i> | : Increase in Soil Organic Carbon Stock between initial and post-harvest sampling (t C ha ⁻¹) |
| <i>pH</i> | : Potential of Hydrogen (Negative logarithm of hydronium ion activity; dimensionless) |
| <i>EC</i> | : Electrical Conductivity (dS m ⁻¹) or deciSiemens per metre) |
| <i>Available N</i> | : Alkaline permanganate-extractable Soil Nitrogen (kg ha ⁻¹) |
| <i>Available P</i> | : Olsen-extractable (0.5 M NaHCO ₃ , pH 8.5) Soil Phosphorus (kg ha ⁻¹) |
| <i>Available K</i> | : Neutral normal ammonium acetate-exchangeable Soil Potassium (kg ha ⁻¹) |
| <i>BD</i> | : Soil Bulk Density (Mg m ⁻³) or Megagrams per cubic metre) |

Agronomic Yield & Statistical Parameters:

| | |
|-------------------------|--|
| <i>RDF</i> | : Recommended Dose of Chemical Fertilizers |
| <i>AMF</i> | : Arbuscular Mycorrhizal Fungi |
| <i>Pods Plant-1</i> | : Total number of filled reproductive pods harvested per individual plant unit |
| <i>1000-Seed Weight</i> | : The absolute weight mass of exactly one thousand individual grains (g) |
| <i>Grain Yield</i> | : Net economic seed harvest mass calculated per unit area (kg ha ⁻¹) |
| <i>Biomass Yield</i> | : Cumulative biological dry matter (grain + stover) per unit area (kg ha ⁻¹) |
| <i>RBD</i> | : Randomized Block Design |
| <i>ANOVA</i> | : Analysis of Variance |
| <i>SEm (±)</i> | : Standard Error of the Mean |
| <i>CD (P=0.05)</i> | : Critical Difference at a 5 % mathematical probability threshold |
| <i>NS</i> | : Non-Significant variation pattern (P > 0.05) |

1. Introduction

Soil organic carbon (SOC) is a key indicator of soil fertility, sustainability, and how productive an ecosystem is. It helps maintain soil structure, nutrient cycles, water retention, microbial activity, and overall soil health (Lal, 2004). However, continuous farming and heavy use of chemical fertilizers have led to a drop in SOC levels, which in turn lowers productivity and harms soil quality. So, finding sustainable ways to boost carbon storage and restore soil fertility is important for long-term farming success (Lehmann, 2007).

Agricultural residues are a major source of organic matter that can improve soil carbon levels if properly reused (Mary & Anitha, 2019; Dash et al., 2025). Sugarcane waste, like trash, bagasse, and press mud, is produced in large amounts by the sugar industry (Tandon et al., 2025). India is one of the biggest sugarcane producers, generating significant amounts of sugarcane waste every year, with nearly 40% going unused (Dash et al.,

2025). These unused residues often cause problems like in-field burning (Tandon et al., 2025). Turning sugarcane waste into biochar and organic amendments is a promising way to manage waste and support sustainable farming (Lehmann & Joseph, 2015; Hariyono et al., 2020).

Biochar is a carbon-rich material made from biomass that is heated under low oxygen conditions (Lehmann & Joseph, 2015). Its porous structure and stable carbon make it useful for improving soil fertility, holding nutrients better, boosting microbial activity, and storing carbon in the soil for longer (Glaser et al., 2002; Agegnehu et al., 2016). Using biochar has been found to improve soil properties in physical, chemical, and biological ways, promoting sustainable farming (Aluthge et al., 2025).

Natural farming uses local organic materials and biological processes to keep soil fertile while reducing the need for synthetic chemicals. Inputs like Jeevamrut, mulching, compost, and crop residues help increase microbial activity and nutrient recycling (Vessey, 2003). Black gram (*Vigna mungo* L.), a pulse crop, responds well to better soil conditions because of its ability to fix nitrogen and its short growing cycle (Jackson, 1973). Though many studies show the benefits of organic amendments on soil and crop growth (Walkley & Black, 1934; Mary & Anitha, 2019), there isn't much information on how combining sugarcane trash biochar with natural farming practices affects SOC storage and nutrient uptake in black gram.

Although several studies have reported the beneficial effects of biochar and organic amendments on soil fertility and crop productivity, limited information is available regarding the combined influence of sugarcane residue-based amendments and natural farming practices on soil carbon sequestration and black gram productivity. Therefore, the present study was undertaken to evaluate the effects of sugarcane residue-derived organic amendments on soil organic carbon, soil physio-chemical properties, nutrient uptake, carbon sequestration efficiency, and yield of black gram under natural farming protocols. The study also aimed to compare these practices with conventional fertilizer management.

2. Materials and Methods

2.1 Experimental Site

The field experiment was conducted during the kharif season at the research farm of Krushi India Corporation Limited, Surat district, Gujarat, India. The experimental site is situated in a semi-arid tropical region characterized by warm climatic conditions and monsoon rainfall. The area receives an average annual rainfall of approximately 1100–1200 mm, with most precipitation occurring during June to September. During the cropping season, the average temperature ranged from 22°C to 37°C.

The experimental soil was sandy loam in texture with low to medium fertility status. Initial soil properties are presented in Table 1. The soil had pH 7.42, electrical conductivity 0.34 dS m⁻¹, organic carbon 0.42%, available nitrogen 228.5 kg ha⁻¹, available phosphorus 24.8 kg ha⁻¹, available potassium 248.2 kg ha⁻¹, and bulk density 1.48 Mg m⁻³.

2.2 Experimental Design and Treatments

Each experimental plot measured 4.0 m × 3.0 m (12 m²). A buffer space of 0.5 m was maintained between plots and 1.0 m between replications to avoid interference among treatments.

The study used a Randomized Block Design with six treatments and three replications. The treatments were:

- **T1:** Control (no input)
- **T2:** Sugarcane trash mulching
- **T3:** Sugarcane bagasse compost
- **T4:** Press mud application
- **T5:** Natural farming practices (Jeevamrut + mulch + biochar)
- **T6:** Recommended chemical fertilizers (RDF)

Each plot was spaced apart to prevent nutrient movement between treatments.

2.3 Crop Establishment

Black gram (*Vigna mungo* L.) was sown following standard agricultural practices. Seeds were planted at appropriate spacing under rain-fed conditions. All treatments had consistent intercultural operations and irrigation management.

2.4 Preparation and Application of Biochar

Sugarcane trash biochar was prepared through slow pyrolysis under limited oxygen conditions. The biochar was crushed into smaller particles before field application. Organic amendments were applied before sowing and thoroughly incorporated into the soil.

Sugarcane trash mulch, bagasse compost, and press mud were applied according to their respective organic carbon contribution levels. Jeevamrut was applied periodically as per natural farming recommendations. The natural farming treatment (T₅) consisted of the combined application of biochar, Jeevamrut, and mulching materials. Since biochar was evaluated as a component of the natural farming package, the study primarily assessed the integrated effect of biochar-based natural farming rather than the independent effect of biochar alone (Tandon et al., 2025).

2.5 Soil Sampling and Analysis

Soil samples were collected from each plot before planting and after harvest from a depth of 0–15 cm. They were air-dried, processed, and tested for the following:

- Soil organic carbon (Walkley and Black method) (Walkley & Black, 1934)
- Soil pH and electrical conductivity (Jackson, 1973)
- Available nitrogen (Kjeldahl method) (Jackson, 1973)
- Available phosphorus (Olsen's method) (Jackson, 1973)
- Available potassium (Flame photometric method) (Jackson, 1973)
- Bulk density (Core method) (Jackson, 1973)

2.6 Plant Analysis

Plant samples collected at harvest were dried in an oven and tested for nutrient content.

- Nitrogen was estimated using the Kjeldahl method (Jackson, 1973).
- Phosphorus was determined using spectrophotometry (Jackson, 1973).
- Potassium was measured using a flame photometer (Jackson, 1973)

2.7 Soil Organic Carbon Stock and Carbon Accumulation

Soil organic carbon stock was calculated using soil organic carbon concentration, bulk density, and sampling depth (0–15 cm) according to the following equation:

$$\text{SOC Stock (t C ha}^{-1}\text{)} = \text{SOC (\%)} \times \text{Bulk Density (Mg m}^{-3}\text{)} \times \text{Soil Depth (m)} \times 100$$

Carbon accumulation was calculated as the difference between post-harvest SOC stock and initial SOC stock.

2.8 Statistical Analysis

The experimental data was analysed with standard ANOVA methods suitable for a Randomized Block Design. To check if treatments had a real effect, an F-test was used at a 5% significance level ($p \leq 0.05$). Critical Difference (CD) values were calculated to compare the average results when the F-test showed significant differences.

3. Results and Discussion

3.1 Effect on Soil Organic Carbon

The treatments had a strong effect on soil organic carbon. The highest level of SOC, 0.68%, was seen in the treatment with natural farming and biochar (T₅), which was much better than the control (0.41%). Press mud

application (T4) also performed well with SOC of 0.64%. The increase in SOC was because of adding stable carbon and helping microbes break down organic matter more effectively.

Biochar helps store carbon long-term because its carbon is hard for microbes to break down. Adding sugarcane residues helped increase microbial activity and build up organic matter in the soil.

Table 1. Initial soil properties of experimental field

| Parameter | Value | Method |
|---|------------|----------------------|
| Soil texture | Sandy loam | Hydrometer method |
| pH | 7.42 | Digital pH meter |
| EC (dS m ⁻¹) | 0.34 | Conductivity meter |
| Organic Carbon (%) | 0.42 | Walkley–Black method |
| Available Nitrogen (kg ha ⁻¹) | 228.5 | Subbiah & Asija |
| Available Phosphorus (kg ha ⁻¹) | 24.8 | Olsen’s method |
| Available Potassium (kg ha ⁻¹) | 248.2 | Flame photometer |
| Bulk Density (Mg m ⁻³) | 1.48 | Core method |

3.2 Effect on Soil Physical and Chemical Properties

The post-harvest soil properties as influenced by different treatments are presented in Table 2 and illustrated in Fig. 1. Significant improvements in soil organic carbon, nutrient availability, and bulk density were observed under sugarcane residue-based amendments compared with the control treatment.

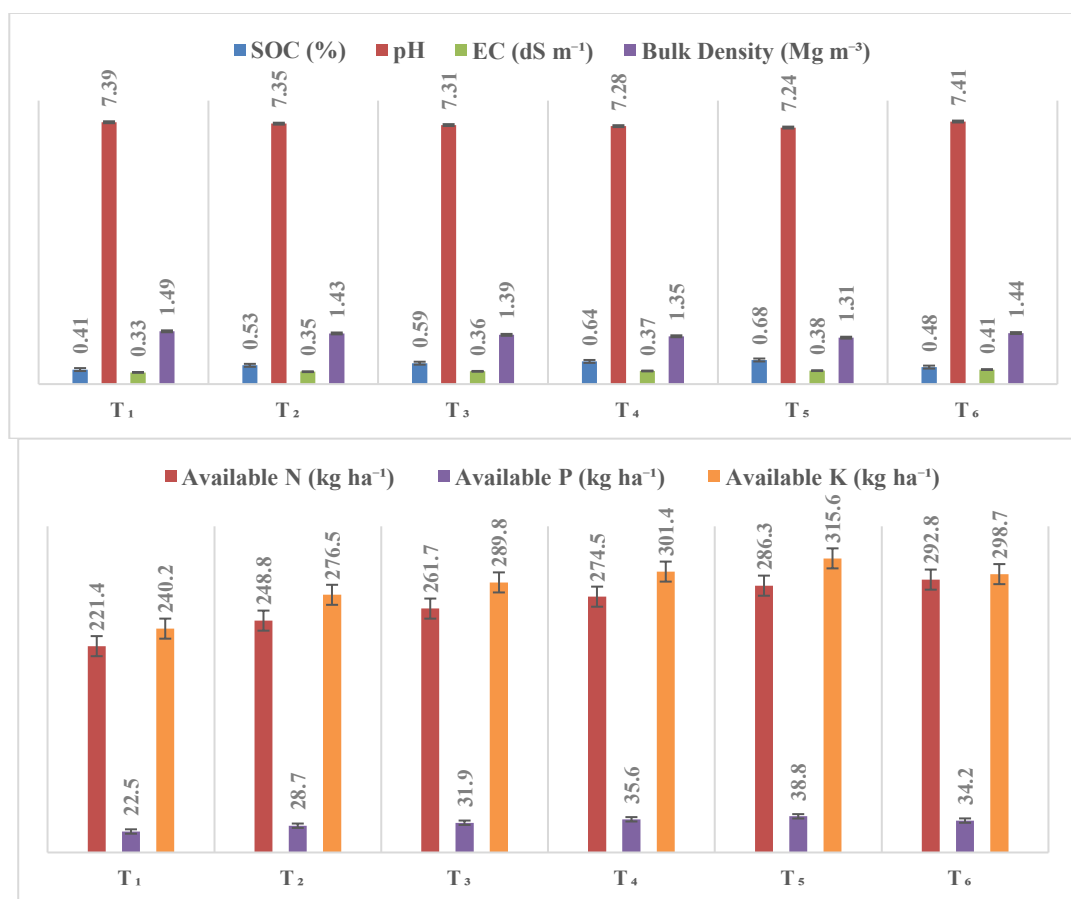


Fig. 1. Effect of sugarcane trash biochar and organic amendments on soil organic carbon content after harvest

Table 2. Effect of sugarcane trash biochar and organic amendments on soil properties after harvest

| Treatment | SOC (%) | pH | EC (dS m ⁻¹) | Available N (kg ha ⁻¹) | Available P (kg ha ⁻¹) | Available K (kg ha ⁻¹) | Bulk Density (Mg m ⁻³) |
|--|---------|------|--------------------------|------------------------------------|------------------------------------|------------------------------------|------------------------------------|
| T ₁ – Control | 0.41 | 7.39 | 0.33 | 221.4 | 22.5 | 240.2 | 1.49 |
| T ₂ – Trash Mulching | 0.53 | 7.35 | 0.35 | 248.8 | 28.7 | 276.5 | 1.43 |
| T ₃ – Bagasse Compost | 0.59 | 7.31 | 0.36 | 261.7 | 31.9 | 289.8 | 1.39 |
| T ₄ – Press Mud | 0.64 | 7.28 | 0.37 | 274.5 | 35.6 | 301.4 | 1.35 |
| T ₅ – Natural Farming + Biochar | 0.68 | 7.24 | 0.38 | 286.3 | 38.8 | 315.6 | 1.31 |
| T ₆ – RDF | 0.48 | 7.41 | 0.41 | 292.8 | 34.2 | 298.7 | 1.44 |
| SEm (±) | 0.02 | 0.04 | 0.01 | 5.21 | 1.12 | 4.85 | 0.03 |
| CD (P=0.05) | 0.05 | NS | 0.03 | 15.62 | 3.35 | 14.54 | 0.08 |

NS = non-significant

Key Trends in Post-Harvest Soil Properties:

Soil Organic Carbon (SOC): Different treatments had a big effect on SOC levels. The highest SOC, at 0.68%, was found in the natural farming system combined with biochar (T₅), which was much higher than the control (0.41%). The next highest was press mud application (T₄), with SOC at 0.64%.

Nutrient Availability: T₅ had the most available phosphorus at 38.8 kg per hectare and the most available potassium at 315.6 kg per hectare. Available nitrogen was highest in T₆ (RDF) at 292.8 kg per hectare, which was similar to T₅'s 286.3 kg per hectare.

Physical Architecture: Adding organic material helped reduce soil bulk density and improved the soil's ability to hold water. The T₅ plots had the lowest bulk density at 1.31 Mg per cubic meter, which boosted soil porosity because of the porous nature of biochar particles (16).

3.3 Nutrient Uptake by Black Gram

The effect of treatments on nutrient uptake by black gram is presented in Table 3 and Fig. 2. The highest nitrogen uptake (72.4 kg ha⁻¹), phosphorus uptake (15.6 kg ha⁻¹), and potassium uptake (51.8 kg ha⁻¹) were recorded under natural farming integrated with biochar (T₅). This treatment significantly outperformed the control and was statistically comparable with RDF (T₆) for certain parameters. Enhanced nutrient uptake under T₅ may be attributed to improved nutrient availability, increased microbial activity, and better root development resulting from the combined effects of biochar and natural farming inputs (12).

Table 3. Effect of treatments on nutrient uptake by black gram

| Treatment | Nitrogen Uptake (kg ha ⁻¹) | Phosphorus Uptake (kg ha ⁻¹) | Potassium Uptake (kg ha ⁻¹) |
|--|--|--|---|
| T ₁ – Control | 42.8 | 8.2 | 28.6 |
| T ₂ – Trash Mulching | 54.3 | 10.5 | 36.8 |
| T ₃ – Bagasse Compost | 61.7 | 12.4 | 42.3 |
| T ₄ – Press Mud | 67.8 | 14.2 | 46.5 |
| T ₅ – Natural Farming + Biochar | 72.4 | 15.6 | 51.8 |
| T ₆ – RDF | 69.5 | 13.8 | 48.1 |
| SEm (±) | 1.86 | 0.48 | 1.74 |
| CD (P=0.05) | 5.58 | 1.43 | 5.21 |

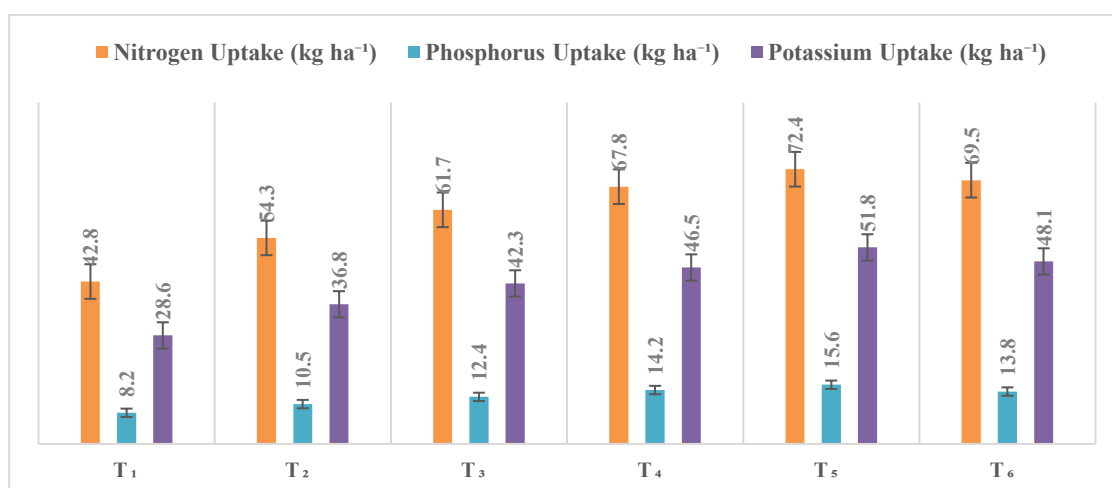


Fig. 2. Influence of treatments on nutrient uptake (N, P, and K) by black gram

3.4 Effect on Grain Yield and Biomass

Yield attributes and yield performance of black gram under different treatments are presented in Table 4 and Fig. 3. Significantly higher grain yield (1328 kg ha⁻¹) and biomass yield (3025 kg ha⁻¹) were recorded under natural farming integrated with biochar (T₅), followed by RDF (1265 kg ha⁻¹) and press mud application (1216 kg ha⁻¹). The superior performance of T₅ may be attributed to improved soil fertility, enhanced nutrient retention, greater microbial activity, and better moisture conservation resulting from the combined action of biochar, mulching, and biological inputs (13, 16).

Table 4. Effect of treatments on yield and yield attributes of black gram

| Treatment | Plant Height (cm) | Pods Plant ⁻¹ | 1000-Seed Weight (g) | Grain Yield (kg ha ⁻¹) | Biomass Yield (kg ha ⁻¹) |
|--|-------------------|--------------------------|----------------------|------------------------------------|--------------------------------------|
| T ₁ – Control | 32.4 | 18.6 | 36.2 | 712 | 1845 |
| T ₂ – Trash Mulching | 38.7 | 24.2 | 38.5 | 926 | 2264 |
| T ₃ – Bagasse Compost | 42.1 | 27.8 | 39.6 | 1084 | 2518 |
| T ₄ – Press Mud | 45.5 | 31.6 | 40.8 | 1216 | 2794 |
| T ₅ – Natural Farming + Biochar | 48.2 | 34.5 | 42.1 | 1328 | 3025 |
| T ₆ – RDF | 46.4 | 32.8 | 41.2 | 1265 | 2886 |
| SEm (±) | 1.12 | 1.45 | 0.76 | 42.8 | 78.4 |
| CD (P=0.05) | 3.35 | 4.32 | 2.28 | 128.4 | 235.1 |



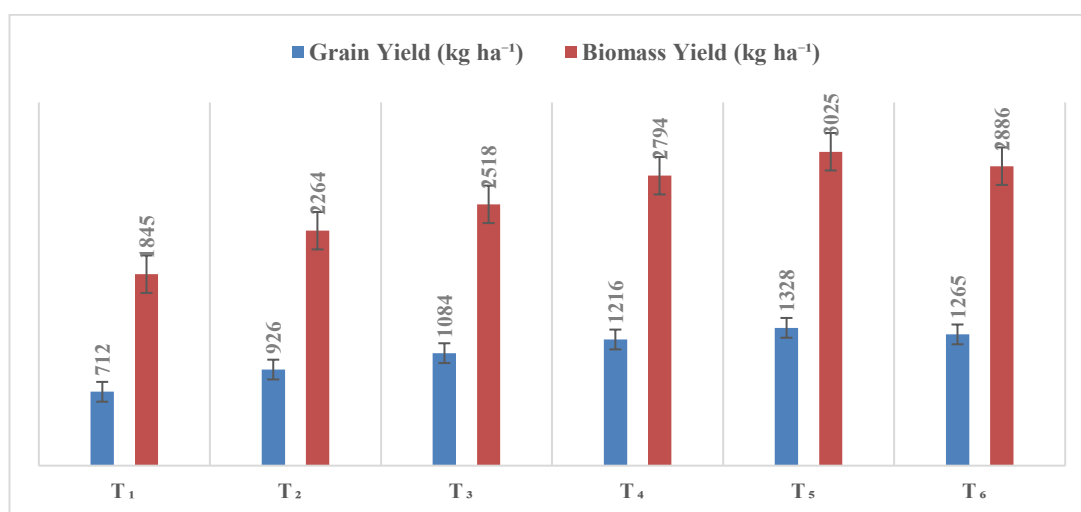


Fig. 3. Effect of biochar and natural farming practices on grain yield of black gram

3.5 Soil Organic Carbon Stock and Carbon Accumulation

Soil Organic Carbon Stock and Carbon Accumulation under Different Organic Amendment Treatments is presented in Table 5 and Fig. 4.

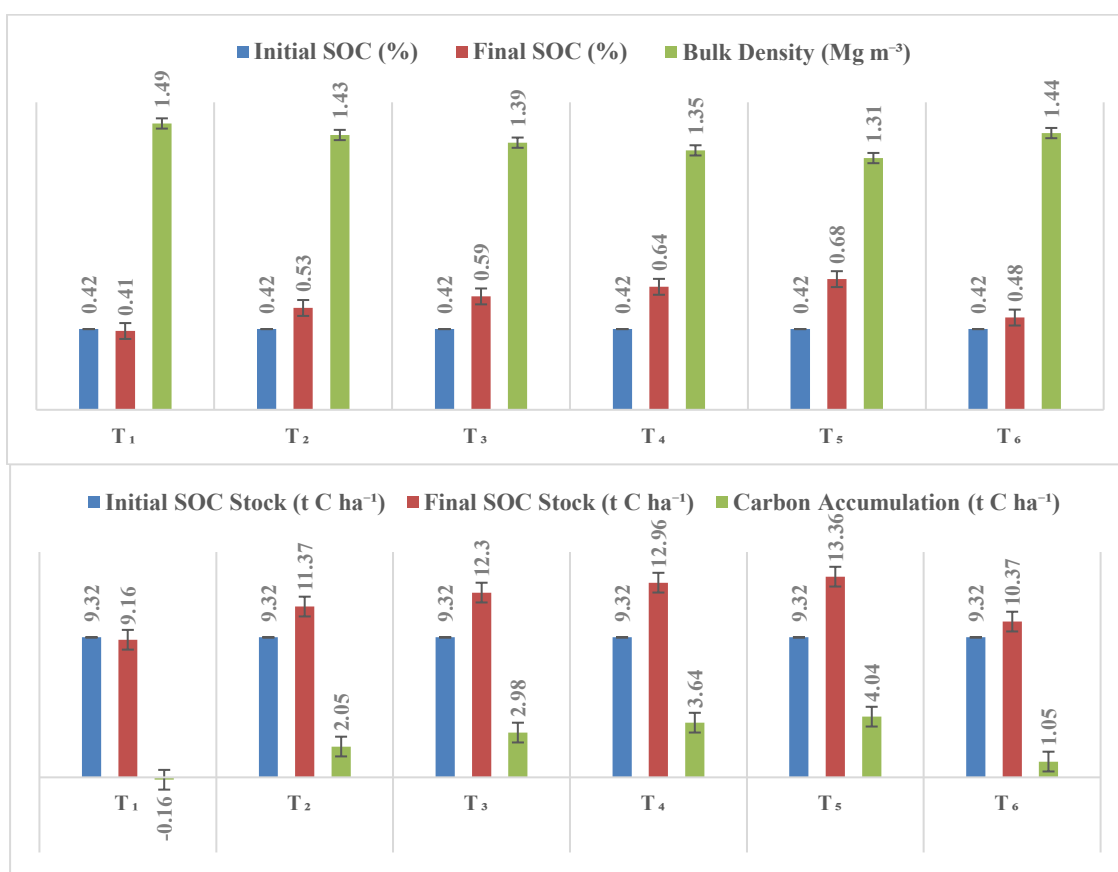


Fig. 4. Soil organic carbon stock and carbon accumulation under different organic amendment treatments

Soil organic carbon stock increased significantly under all organic amendment treatments compared with the control. The highest post-harvest SOC stock (13.36 t C ha⁻¹) and carbon accumulation (4.04 t C ha⁻¹) were recorded under natural farming integrated with biochar (T₅), followed by press mud application (12.96 t C ha⁻¹ and 3.64 t C ha⁻¹, respectively). The control treatment showed a slight decline in SOC stock, indicating depletion of native soil carbon in the absence of external organic inputs. These findings demonstrate the potential of sugarcane residue-based amendments for enhancing soil carbon storage and improving soil health (Lal, 2004).

Table 5. Soil organic carbon stock and carbon accumulation under different organic amendment treatments

| Treatment | Initial SOC (%) | Final SOC (%) | Bulk Density (Mg m ⁻³) | Initial SOC Stock (t C ha ⁻¹) | Final SOC Stock (t C ha ⁻¹) | Carbon Accumulation (t C ha ⁻¹) |
|--|-----------------|---------------|------------------------------------|---|---|---|
| T ₁ – Control | 0.42 | 0.41 | 1.49 | 9.32 | 9.16 | -0.16 |
| T ₂ – Trash Mulching | 0.42 | 0.53 | 1.43 | 9.32 | 11.37 | 2.05 |
| T ₃ – Bagasse Compost | 0.42 | 0.59 | 1.39 | 9.32 | 12.3 | 2.98 |
| T ₄ – Press Mud | 0.42 | 0.64 | 1.35 | 9.32 | 12.96 | 3.64 |
| T ₅ – Natural Farming + Biochar | 0.42 | 0.68 | 1.31 | 9.32 | 13.36 | 4.04 |
| T ₆ – RDF | 0.42 | 0.48 | 1.44 | 9.32 | 10.37 | 1.05 |

3.6 Discussion

The study demonstrated that sugarcane residue-based amendments, particularly biochar-integrated natural farming, significantly improved soil quality and black gram productivity. The steady increase in soil organic carbon in biochar treatments comes from its strong, stable aromatic carbon structure, which is hard for microbes to break down (Zafeer et al., 2024). These findings match those of Johannes Lehmann and his team, who saw better carbon storage and soil fertility after applying biochar to tropical soils (Lehmann & Joseph, 2015).

Improving physical soil issues like reduced bulk density and better water holding ability is linked to the large internal pores and surface area of sugarcane trash biochar (Hariyono et al., 2020). This structure creates stable spaces that help roots grow and microbes thrive. Bruno Glaser and others have also noted that using charcoal-based materials greatly improves the physical and chemical conditions of old tropical soils (Glaser et al., 2002).

The higher levels of key nutrients like nitrogen, phosphorus, and potassium in biochar and organic treatments suggest slower nutrient breakdown and less loss through chemical leaching (Mary & Anitha, 2019). Biochar acts like a nutrient trap, keeping mobile nutrients in place (Glaser et al., 2002). At the same time, using liquid biostimulants (Jeevamrut) and biofertilizers with Rhizobium, Azotobacter, and phosphate-solubilizing bacteria speeds up natural nitrogen fixation and nutrient binding (Vessey, 2003; Bhattacharyya & Jha, 2012), matching the results of chemical fertilizers. These combined effects align with Claudia Steiner and her team, who found that using biochar with manure improves nutrient use and raises crop yields in a sustainable way (Steiner et al., 2007).

The superior performance of biochar-integrated natural farming systems may also be attributed to enhanced soil microbial activity and improved nutrient cycling. Biochar provides a favorable habitat for beneficial microorganisms due to its porous structure and large surface area, thereby improving nutrient retention and biological activity within the rhizosphere. Similar observations were reported by Agegnehu et al. (2016), who documented increased nutrient-use efficiency and crop productivity following combined application of biochar and organic amendments. Furthermore, recent studies on sugarcane-derived biochar have demonstrated its potential to improve soil aggregation, moisture retention, and carbon stabilization under tropical agricultural conditions.

The observed increase in soil organic carbon stock and carbon accumulation under biochar and press mud treatments highlights the potential of sugar industry residues as climate-smart soil amendments. These organic materials not only recycle agricultural wastes but also contribute to long-term carbon storage and mitigation of

greenhouse gas emissions. Such practices align with global sustainable development goals aimed at improving soil health while reducing agriculture's environmental footprint.

4. Conclusion

The study showed that sugarcane residue-based amendments improved soil organic carbon, soil nutrient availability, bulk density, nutrient uptake, and yield of black gram compared with the untreated control. Among the evaluated treatments, natural farming with biochar recorded the highest soil organic carbon content, nutrient uptake, grain yield, biomass yield, soil organic carbon stock, and carbon accumulation. The treatment recorded 0.68% soil organic carbon, 1328 kg ha⁻¹ grain yield, 3025 kg ha⁻¹ biomass yield, 13.36 t C ha⁻¹ final soil organic carbon stock, and 4.04 t C ha⁻¹ carbon accumulation. Press mud application also contributed to improved soil organic carbon and carbon storage. The results suggest that sugarcane residues can be effectively recycled as soil amendments for improving soil health and supporting black gram productivity. The integration of biochar with natural farming inputs showed promising results under the conditions of the present study. However, as the biochar treatment was applied as part of an integrated package, the independent contribution of biochar could not be separated. Therefore, the findings should be interpreted as the effect of a combined natural farming and biochar-based amendment system rather than biochar alone.

5. Limitations

The present study was conducted during a single kharif season under the agro-climatic conditions of Surat, Gujarat. Therefore, the results represent short-term treatment effects and may not fully indicate the long-term influence of sugarcane residue-based amendments on soil organic carbon dynamics, nutrient cycling, soil biological activity, and crop productivity. The experiment evaluated biochar as a component of an integrated natural farming treatment that also included Jeevamrut and mulching. As a result, the individual effect of biochar could not be separated from the combined treatment effect. The study was also limited to one crop, one location, and one soil type. Further long-term experiments across different agro-ecological regions, soil conditions, and cropping systems are required to validate the consistency and scalability of these findings. Future studies should also include independent biochar treatments and detailed characterization of organic amendments to improve interpretation and reproducibility.

Disclaimer (Artificial Intelligence)

Author(s) hereby declares that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators have been used during the writing or editing of this manuscript.

Competing Interests

Authors have declared that they have no known competing financial interests or non-financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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