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Empirical observation of natural farming inputs on nitrogen uptake, soil health, and crop yield of rice-wheat cropping system in the organically managed Inceptisol of Trans Gangetic plain

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Natural Farming represents an agro-ecological methodology for farming that emphasizes regenerative practices with an aim to promote holistic ecological balance and reduce the dependence on external inputs as well as financial resources. Substantial concern has recently arisen over the need to promote agroecosystems that are more sustainable in order to improve the deteriorating soil health as well as reversing the yield plateau of crop. So, the current on farm field experiment was executed comprising of 8 treatments with different combination of natural farming inputs (Ghanjeevamrit, Jeevamrit, Beejamrit), organic fertilizer (such as FYM), integrated nutrient management (NPK, FYM, Azotobacter and Azolla) and in-organic(NPK) to examine and compare the consequence of natural farming inputs, organic fertilizer and in-organic dosage of fertilizer on soil nitrogen uptake, soil physicochemical properties, soil biological properties, soil microbial population and crop yields in a rice-wheat cropping system over two crop seasons 2021–23 [rice (Pusa-1509) and wheat (HD-3086)]. The study results demonstrated that there was significant ($p < 0.05$) increase in the soil's nitrogen availability and nitrogen uptake with the use of natural farming inputs as compared to control treatment, whereas, natural farming treatments (T_{NF1} , T_{NF2} , T_{NF3} , T_{MNF}) were inferior than integrated nutrient management (T_{INM}) and recommended doses of fertilizer (T_{RDF}) treatment in case of nitrogen uptake by both rice and wheat crop. The soil enzymatic activity (Dehydrogenase, β -glucosidase, and urease), soil microbial biomass carbon and nitrogen, and soil microbial population (Bacteria, fungi, and actinomycetes) were significantly ($p < 0.05$) higher in treatment receiving natural farming inputs compare to in-organic fertilizer and organic fertilizer. A positive and significant correlation was observed between potential mineralization nitrogen and soil enzymatic activity (Dehydrogenase, β -glucosidase, and urease), soil microbial biomass carbon and nitrogen and soil microbial population (Bacteria, fungi, and actinomycetes). The crop yield at the end of experiment recorded to be highest in treatment T_{INM} (75% RDF (In-organic) + 25% RDF (FYM) + BGA) i.e., (Rice-

4.76 t/ha and Wheat- 5.82 t/ha) compared to T_{RDF} and T_{NF} . A crop yield reduction of 14.2% was observed in treatment receiving natural farming inputs compare to T_{INM} . A significant increase in crop yield was observed in T_{MNF} (Jeevamrit (25%) + Ghanjeevamrit (25%) + 50% RDF through FYM + Beejamrit) compare to T_c (Control) and T_{FYM} (Farmyard manure). Therefore, our study suggests that adoption of natural farming inputs over time can facilitate the enhancement of soil biological health of Inceptisol of Trans Gangetic Plain of India.

KEYWORDS

Ghanjeevamrit, Jeevamrit, Beejamrit, nitrogen uptake, soil microbial population

1 Introduction

Green revolution refers to a period of significant agricultural advancement in India that took place from mid-20th century onwards. As green revolution can be seen as subset of conventional farming practices, it was promoted and implemented on a large scale with the objective to increase food production and to address food shortage. It introduces modern farming techniques and technologies which include use of synthetic fertilizers, pesticides, herbicides and farm mechanization (John and Babu, 2021). Even though green revolution has effectively accomplished its core objective of augmenting agricultural productivity and ensuring food self-sufficiency in the country. But the exclusive dependence on inorganic agrochemical has engendered significant adverse repercussions on the environment. For instance, the widespread usage has resulted in soil degradation, depletion of soil microbiota and environmental pollution (Pingali, 2012). Livestock husbandry and the application of nitrogen-based fertilizers stand as the primary factors responsible for agriculture's direct contribution to 20% of the country's greenhouse gas emissions (Vetter et al., 2017; Anas et al., 2020). According to Craswell (2021), these fertilizers serve as the principal catalyst for nitrate contamination in surface water ecosystems. Land degradation is also underway in more than 30% of the nation's entire geographic area (Kabiraj et al., 2022). The strain from the environment has a damaging impact not only on agriculture output and natural resources but also on human health as well. A significant variation in yield of agricultural crops is observed despite inculcating the best practices and intensive use of farm inputs. Additionally, the quality of agricultural land is declining, which increases the threat to the agro-ecology and soil resources. These perils encompass species extinction, desertification, climate change, as well as soil, air, water, and food supply contamination (Gupta, 2019). Synthetic agro-chemical impact human wellness, because residues from in-organic fertilizer used in crop and soil gets biomagnified and ultimately enter the digestive tracts of individuals who consume these dietary items. As a result, the human body is showing adverse effects on health, including interference of the immune, neurological, and hormonal systems (Nicolopoulou-Stamati et al., 2016). Every nation grappling with issues of poverty, starvation, and undernourishment will be compelled to accelerate agricultural expansion as a means to attain sustainable development goals, particularly as they endeavor to achieve the eradication of poverty, the elimination of hunger, and the establishment of a salubrious environment for all (Paroda, 2018). There is growing fear that any further efforts to maintain this chemical agriculture model will

be futile in the long run and irreparably harm soil health. Agriculture's sustainability is one of the world's top issues right now. Returning to non-chemical agriculture has become crucial to achieving production sustainability. Organic farming is a methodology rooted in the principles and dynamics of living organisms, wherein all constituents, including soil, plants, farm animals, microorganisms, insects, farmers, and others, exhibit interdependence. According to Nagavani and Subbian (2015), using organic manures in conjunction with inorganic fertilizers is frequently seen as the answer to maintaining high levels of productivity while maintaining environmental safety. In the hunt for more farmer-friendly as well as eco-friendly alternative systems of farming. Several environmentalists, among them Subhash Palekar from India, Chao from Korea, and Masanobu Fukuoka from Japan, introduced the concept of natural farming, commonly referred to as zero budget natural farming (ZBNF) in India. Natural farming is an agro-ecological approach to agriculture that promotes the cultivation of plants in symbiosis with their surrounding ecosystem. The two main axes of natural farming are structural and agronomic. It entails the enrichment of soil fertility through the application of diverse agro-ecological principles, including strategies like crop diversification, mulching with crop residues, nutrient recycling, fostering beneficial biotic interactions. Simultaneously, it imposes limitations on the utilization of synthetic fertilizers and external inputs (Palekar, 2006). Contrarily, NF focuses on removing farmers' dependence on outside inputs and financial markets in order to provide them autonomy by forbidding them to make any purchases from outside parties, particularly businesses (Rosset and Martínez-Torres, 2012). Natural farming inputs have a high microbial population, which when used enhances the soil flora and mineralizes the macro- and micronutrients in the soil so that plants can use them and provides a sustainable crop yield. Natural farming experienced a huge surge across the country with numerous states like Andhra Pradesh, Odisha, Himachal Pradesh, Chhattisgarh, Madhya Pradesh etc., adopting natural farming as state policy or a grassroots movement, Khadse et al. (2018). It requires scientific confirmation in terms of its impacts on production in various agro-climatic situations, cropping systems, and soil types in order for multiple state governments to accept natural farming as one of their state policies. It may be best to employ natural farming methods in conjunction with crop diversification along with intercropping to boost crop yield stability, preserve soil health as well as fertility and lower greenhouse gas emissions. With this perspective in consideration, the present investigation was conducted to examine the impact of natural farming on soil quality, nutrient absorption, and crop yield. The primary objective of the study was to.

Evaluate the impact of organic and natural farming inputs (Ghanjeevamrit, Jeevamrit, and Beejamrit) application in comparison with conventional management, on soil health, nitrogen availability and crop yield.

2 Material methods

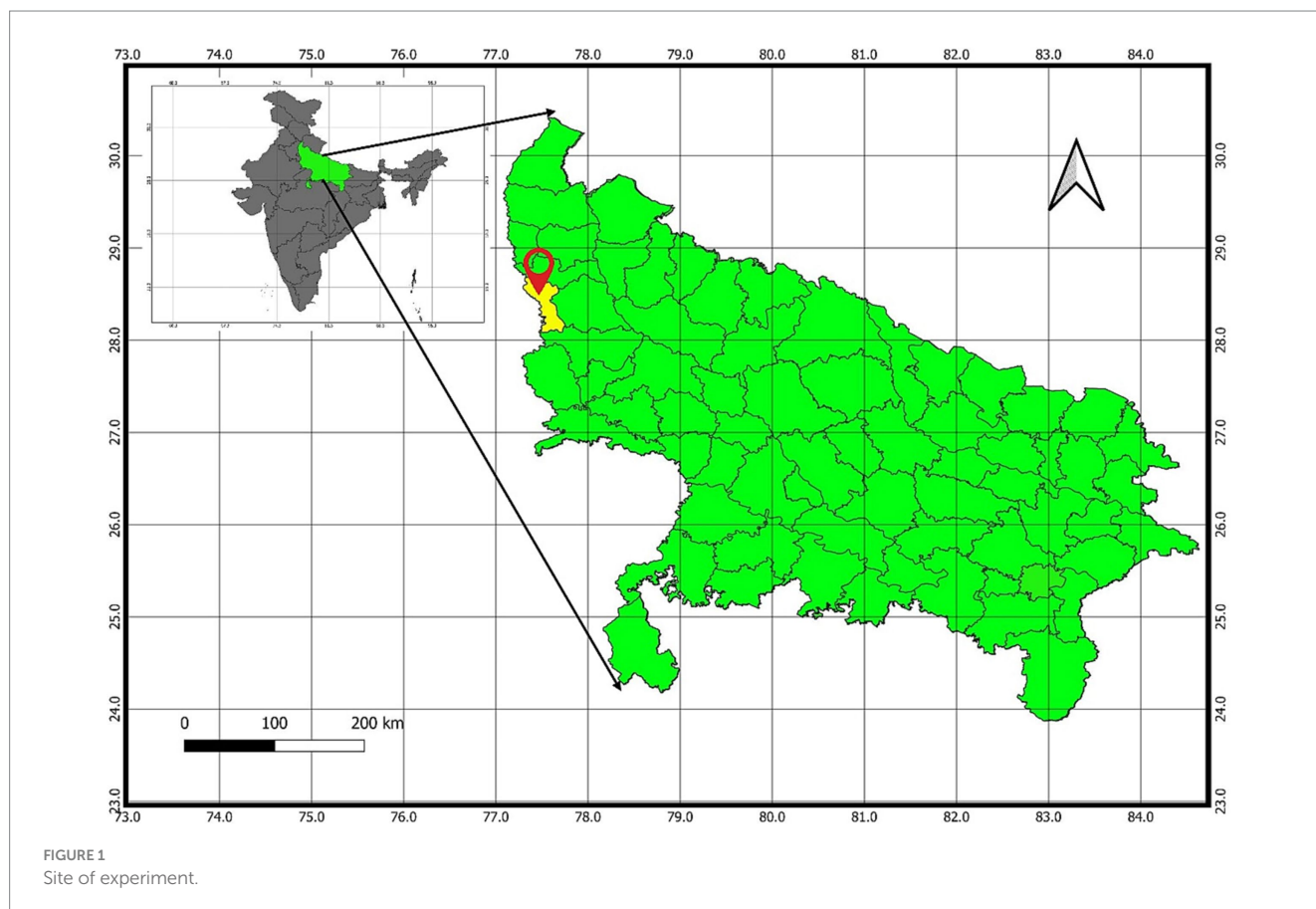
2.1 Experimental site

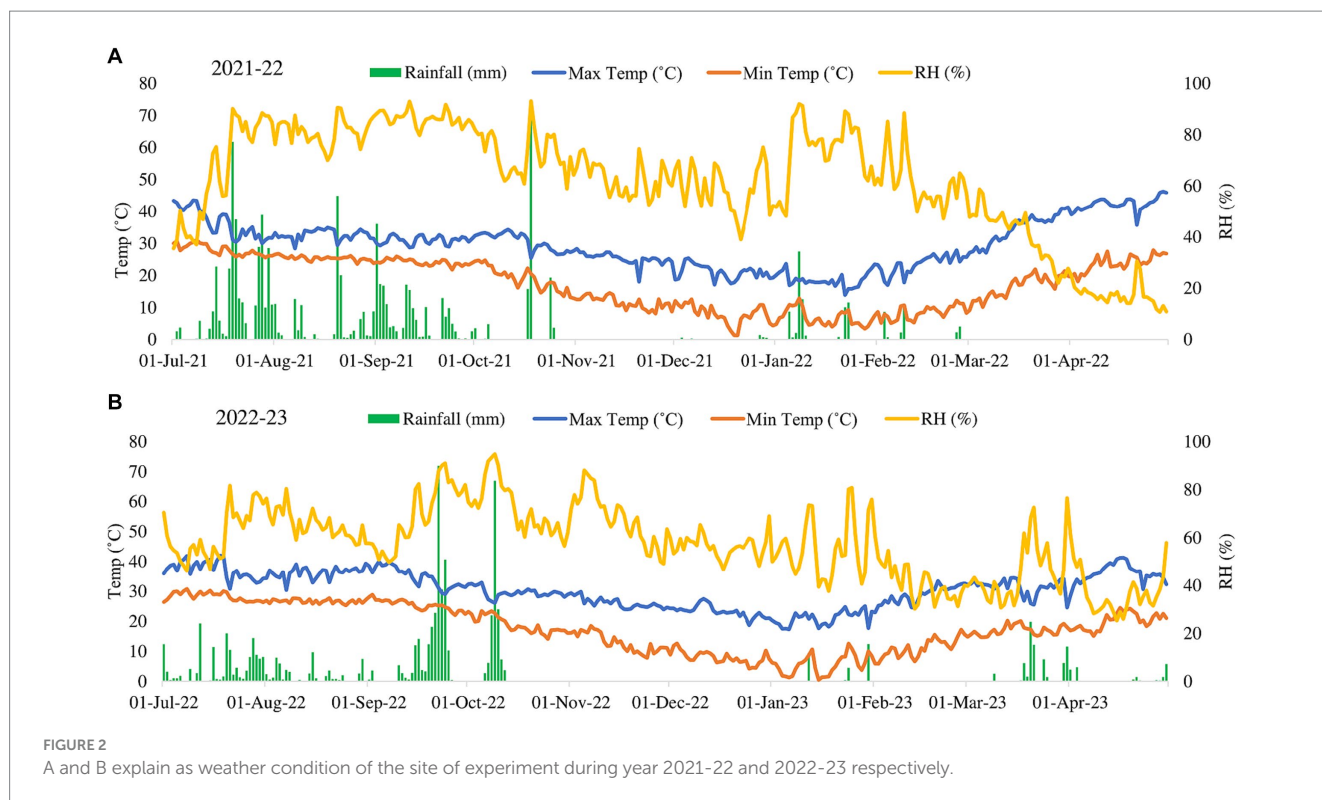
The experiment was undertaken at a farm owned by the farmer (coordinates 28.53° N, 77.64° E) as illustrated in Figure 1, spanning the three-year period from 2021 to 2023. This farm is affiliated with the Indian Council of Agricultural Research (ICAR)-Krishi Vigyan Kendra (KVK), located in Gautambudh Nagar, Uttar Pradesh. The farm is situated at an approximate elevation of 200 m above sea level and experiences an average annual precipitation of 700 mm. The climate in this geographical area falls within the category of sub-tropical and semi-arid, characterized by an annual average temperature of 26 °C. The soil exhibited a sandy clay loam texture, with the following major properties at a depth of 0–20 cm: 46% sand, 33% silt, 20% clay, a pH of 8.3, a bulk density of 1.39 g/cm³, and an organic carbon content of 0.47%. The initial soil nutrient levels, determined prior to seed sowing, were as follows: low available nitrogen (238 ± 0.5 kg/ha), medium levels of available phosphorus (53 ± 0.2 kg/ha), and available potassium (120 ± 0.8 kg/ha). The experiment involved a rice-wheat (*Oryza sativa*-*Triticum aestivum*)

cropping system, utilizing Pusa 1,509 rice and HD-3086 wheat varieties. The experimental trial started with Rice (Kharif season-2021-22) till Wheat (Rabi season-2022-23). Before the initiation of the experimental trial, the site had been under continuous organic cultivation of cereals (rice and wheat) and legumes (mungbean) in rotation for more than 6 years. During the years 2021–22 and 2022–23, the total recorded rainfall was 16.3 mm and 18.7 mm, respectively. The average maximum temperatures during the growing seasons of 2021–22 and 2022–23 were 41.1°C and 42.2°C, while the average minimum temperatures were 10.2°C and 9.8°C, as depicted in Figure 2.

2.2 Experimental design and treatments

The experimental field trial was executed following a randomized block design (RBD) with three replications, encompassing eight distinct treatments, namely T_c (No fertilizer), T_{RDF} (NPK @ 150:60:40 kg/ha), T_{INM} [75% Recommended dose of fertilizer (in-organic) + 25% Recommended dose of fertilizer (FYM) + BGA (Azotobacter for wheat) & (Azolla for rice)], T_{FYM} (Recommended dose of fertilizer through FYM), T_{NF1} (Jeevamrit @ 1,000 L ha⁻¹ in 3 split + Mulching), T_{NF2} (Ghanjeevamrit @ 1,000 kg ha⁻¹ in 3 split + mulching), T_{NF3} (Jeevamrit (50%) + Ghanjeevamrit (50%) in 3 split + Mulching), T_{MNF} (Jeevamrit (25%) + Ghanjeevamrit (25%) + 50% RDN through FYM in 3 split + Beejamrit + mulching). Conventional tillage has been applied from T_c to T_{FYM} and minimum tillage in T_{NF1} to





T_{MNF} . The experimental field, situated within the affiliation of KVK-Gautambudhnagar, Uttar Pradesh. The Control treatment group has deliberately not received any organic treatments, facilitating the establishment of a rigorous and credible basis for comparison with the above designated treatment groups across various parameters. The detailed treatment information has been given in (Table 1). The nitrogen fertilizer, in the form of urea containing 46% nitrogen (N), was applied in three stages: 50% at sowing, 30% during tillering, and 20% at flowering. Phosphorus fertilizer, specifically Di-ammonium phosphate with 46% P_2O_5 , was exclusively applied during the seedling stage. Potassium fertilizer, using potassium chloride with 50% K_2O , was evenly distributed between the seedling and earing stages. The nutrient concentration of these input applications for different treatments is detailed in Table 2. Each experimental plot covered an area of 20 m^2 (5×4).

2.3 Method of soil sample collection and analysis

Fresh soil samples were collected from the 0–15 cm soil layer at three distinct locations within each treatment, employing an 8 cm tube auger. This sampling procedure was conducted during multiple growth stages of the crop, including tillering, flowering, grain filling, and physiological maturity. In total, 24 fresh soil samples were gathered, air-dried for a duration of 7 days, passed through a 2 mm sieve, thoroughly mixed, and stored in plastic bags for subsequent analysis. For soil enzymatic activities, microbial biomass carbon, nitrogen and microbial population analysis fresh soil sample from anthesis stage was collected. The analysis of various soil properties was conducted in accordance with established standard procedures. The

available nitrogen content was determined using the Kjeldahl digestion method (Kjeldahl, 1883). Available soil phosphorus and potassium were assessed using Olsen's method and the ammonium acetate method, respectively (Hanway and Heidel, 1952; Olsen, 1954). Soil organic carbon content was determined through the Walkley and Black Method (Walkley and Black, 1934). The initial soil properties of the experimental field are presented in (Table 3).

2.4 Analysis of plant samples

The nitrogen content in both the grain and straw components was assessed through the Kjeldahl digestion method (Jackson et al., 1973). To estimate nitrogen uptake, the nutrient concentrations, expressed as percentages, were multiplied by the corresponding yields of grain and straw. The total nutrient quantity extracted by the crop was determined by summing the nutrient uptake from both grain and straw, as illustrated in Equation (1).

$$N \text{ Uptake (kg / ha)} = \left[\begin{array}{l} (\text{Grain yield (kg / ha)} \times N \text{ concentration in grain (\%)}) + \\ (\text{Straw yield (kg / ha)} \times N \text{ concentration in straw (\%)}) \end{array} \right] \times 0.01 \quad (1)$$

2.5 Potential nitrogen mineralization

Potential nitrogen mineralization of soil was analyzed by using anaerobic incubation method (Keeney, 1982). Collect the representative soil sample ensuring sufficient volume for analysis. Air dry the sample and pass it through 2 mm sieve the place 100 g of

TABLE 1 Treatment details of the experiment.

Treatment details	Details		
	Inputs		Mulching
	Rice	Wheat	
T _C (Control)	No Fertilizer		----NO----
T _{RDF} (Recommended dosage of fertilizer)	Rice (150:60:40 N: P ₂ O ₅ :K ₂ O) kg/ha	Wheat (120:60:40 N: P ₂ O ₅ :K ₂ O) kg/ha	----NO----
T _{INM} (Integrated nutrient management)	75% RDF (In-organic) + 25% RDF (compost) + BGA (Azolla-300 kg ha ⁻¹ after transplanting of rice)	75% RDF (In-organic) + 25% RDF (compost) + BGA (Azotobacter-100 mL kg ⁻¹ seed)	----NO----
T _{FYM} (Farm yard manure)	RDN through FYM (12,500 kg/ha)	RDN through FYM (10,000 kg/ha)	----NO----
T _{NF-1} (Natural Farming-1)	Jeevamrit @ 1,000 L ha ⁻¹ in 3 split, basal doses of 500 L ha ⁻¹ and 250 L ha ⁻¹ each at tillering and anthesis stage+ Beejamrit (10 L kg ⁻¹ seed treatment)		----YES---- Retention of previous crop residues ✓ In rice (Previous wheat crop residue was retained) ✓ In wheat (previous rice crop residue was retained)
T _{NF-2} (Natural Farming-2)	Ghanjeevamrit @ 1,000 kg ha ⁻¹ in 3 split, basal dose of 500 kg ha ⁻¹ and 250 kg ha ⁻¹ each at tillering and anthesis stage + Beejamrit (10 L kg ⁻¹ seed treatment)		----YES---- Retention of previous crop residues ✓ In rice (previous wheat crop residue was retained) ✓ In wheat (previous rice crop residue was retained)
T _{NF-3} (Natural Farming-3)	Jeevamrit (50%) + Ghanjeevamrit (50%) in 3 split, basal dose, and each at tillering and anthesis stage+ Beejamrit (10 L kg ⁻¹ seed treatment)		----YES---- Retention of previous crop residues ✓ In rice (previous wheat crop residue was retained) ✓ In wheat (previous rice crop residue was retained)
T _{MNF} (Modified natural farming)	Jeevamrit (25%) + Ghanjeevamrit (25%) + 50% RDF through FYM in 3 splits + Beejamrit (10 L kg ⁻¹ seed treatment).		----YES---- Retention of previous crop residues ✓ In rice (previous wheat crop residue was retained) ✓ In wheat (previous rice crop residue was retained)

sample into glass jar and tightly close the lid to create anaerobic condition. The soil samples the incubated at 35°C for 1 week. Collect the soil sample from jar and use Kjeldahl method to measure the mineralized nitrogen in the soil sample.

2.6 Soil microbial properties

2.6.1 Soil biomass carbon

Soil microbial biomass carbon (MBC) was determined in accordance with the protocol delineated by Vance et al. (1987). A soil sample weighing 17.5 grams was collected from each treatment and placed in a hermetically sealed container. One milli liter of chloroform was introduced to the sample, which was subsequently fumigated. Additionally, a non-fumigated set was prepared in a 250 mL flask.

These samples were kept in the dark for 24 h. Following incubation, chloroform was evaporated at 50°C within a Biological Oxygen Demand (BOD) chamber, and the caps were left open for the ensuing 20–24 h. Subsequently, 70 mL of a 0.5 M K₂SO₄ solution was added to these samples, and they were subjected to agitation for 30 min. The supernatant was isolated by filtering the samples through Whatman No. 42 filter paper. The absorbance of the supernatant was promptly assessed at 280 nm for both fumigated and non-fumigated samples. The MBC of the soil samples was then computed using the formula provided below. Equation (2) was employed for calculating the carbon content of the microbial biomass.

$$\text{MBC (mg / kg of soil)} = \frac{\text{OD of fumigated soil} - \text{OD of nonfumigated soil} \times 15487}{\text{Amount of soil used}} \quad (2)$$

TABLE 2 Nutrient concentration of inputs applied to different treatments.

S. No.	Inputs	N (%)	P (%)	K (%)
1	Ghanjeevamrit	1.25	0.98	0.82
2	Jeevamrit	0.85	0.21	0.23
3	Beejamrit	0.46	0.19	0.27
4	Farm yard manure	1.2	0.4	1
5	Urea	46	0	0
6	Diammonium phosphate	18	20.08	0
7	Muriate of potash	0	0	49.5

TABLE 3 Physio-chemical properties of soil before the start of experiment.

Sand %	46
Silt %	30
Clay %	22
Soil textural class	Sandy loam
Bulk density (g cm ⁻³)	1.40
Soil aggregate analysis [Macro-aggregate (0.25–2 mm) and Micro-aggregate (0.053–0.25 mm)] (g 100g ⁻¹)	37.5 g & 33.4
Soil temperature	32°C ± 5
Soil moisture % (field capacity)	33.73
Soil pH (1:2: soil: water)	8.15
Electrical conductivity (dSm ⁻¹)	0.23
Organic carbon (%)	0.43
Soil available N (kg ha ⁻¹)	210
Soil available P (kg ha ⁻¹)	53
Soil available K (kg ha ⁻¹)	132
Soil Sulfur (mg kg ⁻¹)	48

2.6.2 Soil biomass nitrogen

The method proposed by Vance et al. (1987) is widely acknowledged and frequently employed for the expeditious retrieval and measurement of microbial biomass nitrogen in soil. In the experimental procedure, representative soil samples are obtained from each treatment plot. Approximately 10 g of soil is weighed into a fumigation chamber, followed by the addition of 30 mL of chloroform. The chamber is sealed, and thorough shaking ensures even distribution of chloroform. Subsequently, the soil undergoes chloroform fumigation for 24–48 h, leading to the effective termination of microbial biomass. A distinct portion of the soil sample is preserved without fumigation, serving as a non-fumigated control. Following fumigation, nitrogen extraction from both fumigated and non-fumigated samples is carried out using a suitable extractant solution, such as 0.5 M K₂SO₄. Vigorous shaking is employed to facilitate the extraction of nitrogen. The nitrogen concentration in the extracts is then determined using the Kjeldahl digestion method. The calculation of Microbial Biomass Nitrogen (MBN) for the soil samples is executed using a provided formula, with Equation (3) specifically utilized for computing the nitrogen content of the microbial biomass. This systematic approach ensures a comprehensive evaluation of microbial biomass nitrogen in the soil.

$$\text{MBC (mg / kg of soil)} = \text{Nitrogen of fumigated soil} - \text{Nitrogen of non - fumigated soil} \times 0.45 \quad (3)$$

2.6.3 Soil enzymatic activity

Soil samples for enzymatic analysis were collected at the anthesis stage of both the rice and wheat crops. The freshly obtained soil samples were promptly sifted through a 2 mm sieve. Subsamples were extracted for moisture content evaluation, which was determined by measuring the loss in weight after subjecting the subsample of soil to drying at 105°C for a period of 48 h. The assessment of enzymatic activity was performed on all treatment samples in triplicate, with an additional sample serving as the control. The enzymes under investigation included dehydrogenase, β-glucosidase, and urease, and their quantification was carried out through colorimetric analysis of reaction products resulting from the incubation of samples with the appropriate substrate under standardized conditions. The dehydrogenase activity was assessed following the procedure proposed by Casida et al. (1964). This enzymatic activity was determined by measuring the production of triphenyl formazan from triphenyl tetrazolium chloride, which served as a hydrogen acceptor. In this method, 1 gram of air-dried soil was carefully weighed and placed in a 15 mL screw cap tube. Three replicates were maintained for each experimental treatment. To these tubes, 0.2 mL of a 3% w/v solution of 2, 3, 5 triphenyl tetrazolium chloride was added. To ensure a thin layer of water on the soil surface, 0.5 mL of a 1% glucose solution was included. The tubes were then incubated at a controlled temperature of 30 ± 1°C. After 24 h of incubation, 10 mL of methanol was introduced, and the tubes were manually shaken for 1 min. Subsequently, these tubes were left in the dark for 6 h. The developed color intensity was quantified using a spectrophotometer set at 485 nm. Dehydrogenase activity was expressed as milligrams of Triphenyl formazan (TPF) produced per hour per gram of air-dry soil. A standard curve was constructed within a range of 0.005 mg TPF to 0.4 mg per 10 mL of methanol.

β-Glucosidase activity was assessed following the protocol outlined by Eivazi and Tabatabai (1988). In this procedure, Modified Universal Buffer (MUB), toluene, and p-nitrophenyl β-D-glucoside solution each of 4 mL, 0.2 mL and 1 mL were incubated, respectively, with 1 gram of soil at 37°C for 1 h. Subsequently, 0.5 M CaCl₂ and 0.1 M THAM (Tris hydroxy methyl amino methane) each of 1 mL and 4 mL, respectively, were added, and the resulting soil solution was filtered using Whatman No. 1 filter paper. The intensity of yellow color solutions was assessed using a spectrophotometer at a wavelength of 490 nm. These measurements were then compared to a calibration curve established from standards that contained varying quantities of p-nitrophenol, specifically 0, 10, 20, 30, 40, and 50 mg per gram of soil, as measured at 490 nm. The comparison was made with respect to the reference readings from the established standards.

The quantification of urease activity in soil entails the assessment of urea hydrolysis by examining the remaining urea content subsequent to the soil's incubation with a urea solution at a temperature of 37°C, following the method introduced by Bremner and Douglas (1971). The amount of urea that remains after a specific incubation period is used as the basis for estimating urease activity. Urea content is determined through a calorimetric method, which involves the chemical reaction of urea with diacetyl monoxime

(DAM) in the presence of Thio-Semi carbazide (TSC), H_3PO_4 , and H_2SO_4 . Subsequently, the intensity of the resulting red coloration was quantified using a spectrophotometer at a wavelength of 527 nm. The calculation of soil urease activity was carried out according to Equation (4) below, where “B” represents the quantity (mg) of urea initially added, “A” signifies the amount (mg) of urea remaining after the time “t,” and “x” corresponds to the oven-dried quantity (in grams) of soil used during incubation. When time is measured in hours, urease activity can be expressed as milligrams of urea per gram of soil (mg urea g^{-1} soil).

$$\text{Urease activity} = (B-A) \cdot x \cdot t \quad (4)$$

The Geometric Mean (GMenz) of enzyme activity amalgamates data from all enzymes, computed for each treatment, employing the subsequent Equation (5), as previously described by Hinojosa et al. (2004).

$$GM_{enz} = (\text{Dehydrogenase} \times \beta\text{-glucosidase} \times \text{Urease})^{1/3} \quad (5)$$

2.6.4 Culturable soil microbial population

The quantification of viable microorganisms in rhizospheric soils was performed out using the serial dilution method, and the values were expressed as colony forming units (CFUs) per gram of soil, as described by Wollum et al., (1994). Rhizospheric soil samples were systematically diluted by mixing 10 g of soil with 90 mL of sterilized distilled water, with subsequent dilutions up to 10^{-8} . For the quantification of bacteria, fungi, and actinobacteria, nutrient agar (comprising Peptone, Yeast Extract, Sodium Chloride, Agar) each added 5g, 2g, 5g 15g, respectively, and add sufficient distilled water to make up the volume by 1,000 mL, Potato Dextrose Agar (containing Potatoes, Dextrose, Agar) added 200g, 20g and 15g respectively, and Actinomycetes isolation agar (with Sodium caseinate 2.0, L-Asparagine 0.10, Sodium propionate 4.0, Dipotassium phosphate 0.5, Magnesium sulfate 0.1, Ferrous sulfate 0.001, Agar 15.0) media from Himedia® were employed, respectively. Subsequently, the soil suspensions from the 10^{-5} , 10^{-4} , and 10^{-3} dilutions were spread onto petri plates in triplicates containing the respective media for bacterial, fungal, and actinobacterial counts. Bacterial, fungal, and actinomycetes colonies were enumerated after 1, 3–4, and 6–7 days of incubation at $28 \pm 2^\circ\text{C}$, respectively.

2.7 Crop yield

The rice and wheat crops were harvested in October and April, respectively, and subsequently threshed to quantify their grain and straw yields per hectare, with moisture content adjusted to 12%.

2.8 Statistical analysis

The measurements acquired from the experimental study underwent analysis using OPSTAT Software, as described by Sheoran et al. (1998), to compute ANOVA. Subsequently, means were

differentiated through Duncan’s multiple range test (DMRT) at a significance level of $p=0.05$. Each variable’s data underwent assessment using a variance protocol analysis within a randomized block design, subject to verification for statistical significance via the “F” test, following the approach outlined by Gomez and Gomez (1984). The evaluation of the impact of different treatments on crop yield and soil microbial population was carried out using R software, version 4.3.1, as specified by R Development Core Team (2012). The standard error of means (SEm) and the least significant difference (LSD) were computed at a significance level of 5%. The correlation panel matrix was constructed using the “ggplot2” and “corrplot” packages within the R software, version 4.3.1.

3 Results

3.1 Soil chemical properties (pH, electrical conductivity, and soil organic carbon)

The results of the analysis of selected soil chemical properties are presented in (Table 4). Following the harvest of rice and wheat, soil pH and electrical conductivity (EC) exhibited no significant variations when different natural farming inputs were applied. However, a slight decrease in pH was observed in the T_{RDF} treatment. Conversely, there was a marginal increase in these values across all other treatments over the years from 2021 to 2023. Notably, the application of various natural farming inputs significantly influenced the organic carbon content of the soil after the crop harvest. Specifically, the T_{NF3} treatment exhibited a significantly higher soil organic carbon content, measuring 0.51 and 0.58% for the years 2021–22 and 2022–23, respectively. This was on par with the T_{MNF} treatment, followed by T_{NF1} and T_{NF2} , which recorded values of 0.55 and 0.56%, respectively, at the end of the experiment. These values were statistically similar. In contrast, the control treatment displayed the lowest organic carbon content, with measurements of 0.40 and 0.42% for the years 2021–22 and 2022–23, respectively.

3.2 Soil available nitrogen, phosphorus, and potassium

During both the agricultural seasons (2021–22 and 2022–23) of the rice-wheat cropping system, it was observed a noteworthy alteration in the levels of available nitrogen subsequent to the harvest of both rice and wheat crops. The treatment designated as “ T_{INM} ” consistently exhibited the highest recorded levels of available nitrogen, registering values of 234.9 and 241.3 kg per ha^{-1} after the wheat harvest for the respective years 2021–22 and 2022–23. In contrast, the treatments denoted as T_{NF1} , T_{NF2} , and T_{NF3} consistently displayed significantly lower concentrations of available nitrogen following the wheat harvest, in comparison to the T_{INM} treatment. Among the various natural farming inputs employed, the “ T_{MNF} ” treatment consistently demonstrated the highest levels of available nitrogen for both years, with values of 229.3 and 237.8 kg ha^{-1} after the wheat harvest for the years 2021–22 and 2022–23, respectively. Specifically, the T_{INM} treatment exhibited a percentage increase in soil-available nitrogen of 2.42 and 1.47% compared to T_{MNF} after the wheat harvest for the years 2021–22 and 2022–23, respectively. It is noteworthy that

TABLE 4 Effect of different natural farming inputs (Ghanjeevamrit, Jeevamrit, and Beejamrit), organic (FYM) and in-organic fertilizer on soil chemical properties at harvest of wheat crop of 2021–22 and 2022–23.

Treatments	pH		EC (dSm ⁻¹)		OC (%)	
	2021–22	2022–23	2021–22	2022–23	2021–22	2022–23
T _c	8.18	8.20	0.23	0.23	0.40	0.42
T _{RDF}	8.12	8.03	0.25	0.25	0.43	0.46
T _{INM}	8.20	8.24	0.24	0.24	0.45	0.49
T _{FYM}	8.22	8.25	0.24	0.23	0.46	0.51
T _{NF1}	8.25	8.30	0.24	0.25	0.48	0.55
T _{NF2}	8.32	8.35	0.23	0.24	0.49	0.56
T _{NF3}	8.35	8.37	0.23	0.23	0.51	0.58
T _{MNF}	8.32	8.36	0.24	0.24	0.50	0.57
Initial	8.15		0.23		0.43	
SEm	0.08	0.07	0.01	0.01	0.004	0.005
LSD (0.05)	NS	NS	NS	NS	0.012	0.014

T_c, Control; T_{RDF}, Recommended dosage of fertilizer; T_{INM}, Integrated nutrient management; T_{FYM}, Farm yard manure; T_{NF-1}, Natural Farming-1; T_{NF-2}, Natural Farming-2; T_{NF-3}, Natural Farming-3; T_{MNF}, Modified Natural Farming. Brief details of the treatment is given in Table 1.

the “T_{RDF}” treatment consistently exhibited soil-available nitrogen values statistically comparable to those of the T_{INM} treatment following both seasons of the rice-wheat cropping system. Conversely, the control treatment (T_c) consistently displayed the significantly lowest levels of available nitrogen. In comparison to T_{INM}, T_{INM} exhibited a percentage increase of 61.6 and 72.2% in soil-available nitrogen following the wheat harvest for the years 2021–22 and 2022–23, respectively (Figure 3).

The available phosphorus content in the soil exhibited notable enhancements in treatments incorporating natural farming inputs, i.e., T_{NF1}, T_{NF2}, T_{NF3}, and T_{MNF}. In particular, the application of T_{MNF} resulted in significantly elevated levels of available phosphorus after 2 years, amounting to 64.6 and 68.3 kg ha⁻¹ after the wheat harvest for the years 2021–22 and 2022–23, respectively. These values were statistically akin to those observed in the T_{RDF} and T_{INM} treatments. Conversely, the T_{FYM} treatment displayed lower levels of available soil phosphorus for both years, post-wheat harvest, measuring 54.8 and 58.5 kg ha⁻¹ in comparison to the other treatment regimens. During the years 2021–22 and 2022–23, T_{NF1} and T_{NF2} exhibited no statistically significant differences in soil phosphorus content after 2 years of crop harvest. The control treatment consistently manifested substantially reduced levels of available phosphorus, with values of 47.2 and 46.3 kg ha⁻¹ after the wheat harvest, representing a reduction of 36.8 and 47.5% compared to T_{MNF} for the respective years (Figure 4).

Diverse treatment approaches exerted significant influence on the soil's available potassium content, as indicated in Figure 5. In the years 2021–22 and 2022–23, notably elevated levels of available potassium following the rice harvest were documented in the T_{INM} treatment, registering at 135.6 and 139 kg ha⁻¹ after the wheat harvest. These values demonstrated statistical similarity to those obtained in the T_{RDF} treatment, followed closely by T_{MNF}. T_{NF3} displayed levels statistically akin to T_{NF1} and T_{NF2} for both years, with available potassium quantities of 126.4 and 123.2 kg ha⁻¹ following the wheat harvest for the years 2021–22 and 2022–23, respectively. In contrast, the control treatments consistently exhibited the most markedly reduced levels of available potassium, measuring 119.2 kg ha⁻¹ after the wheat harvest in the second year of the crop.

3.3 Potential nitrogen mineralization

The field management practices, including fertilization and crop rotation, had a significant impact on soil mineralized nitrogen, as depicted in Figure 6. The potential nitrogen mineralization exhibited extensive variation across different treatment groups and throughout the duration of the experiment. Relative to the control group (T_c), all other treatment groups demonstrated a noteworthy increase in potential nitrogen mineralization. The highest potential nitrogen mineralization, recorded after the 2022–23 wheat harvest, was observed in the T_{MNF} treatment, with a value of 12.20 mg/kg. This represented a substantial 60.5% increase compared to the T_{RDF} treatment, which had a potential nitrogen mineralization of 7.60 mg/kg. The T_{INM} treatment exhibited a nitrogen mineralization value of 9.10 mg/kg. Among the treatments that received natural farming inputs (T_{NF1}, T_{NF2}, and T_{NF3}), all showed statistically equivalent values in terms of potential nitrogen mineralization. In contrast, the lowest nitrogen mineralization was recorded in the control group, T_c, with a value of 4.5 mg/kg. This value was two-fold lower when compared to the T_{MNF} treatment.

3.4 Nitrogen uptake

The nitrogen uptake by both the rice and wheat crops exhibited considerable variation among the treatment groups across multiple years, as summarized in Figure 7. Nitrogen uptake from the soil reached its maximum value in the T_{INM} treatment for both cropping seasons, regardless of the crop type. In the 2021–22 and 2022–23 seasons, T_{INM} demonstrated the highest nitrogen uptake for both rice and wheat, with values of 124.09 and 127.57 kg ha⁻¹ for rice and 160.19 and 170.98 kg ha⁻¹ for wheat, respectively. Following T_{INM}, T_{RDF} recorded the second-highest nitrogen uptake, with values of 108.19 and 114.42 kg ha⁻¹ for rice and 150.78 and 158.18 kg ha⁻¹ for wheat in the same respective seasons. Notably, there were no statistically significant differences in nitrogen uptake between treatments T_{NF1} and T_{NF3}, as well as between T_{INM} and T_{NF2}. The

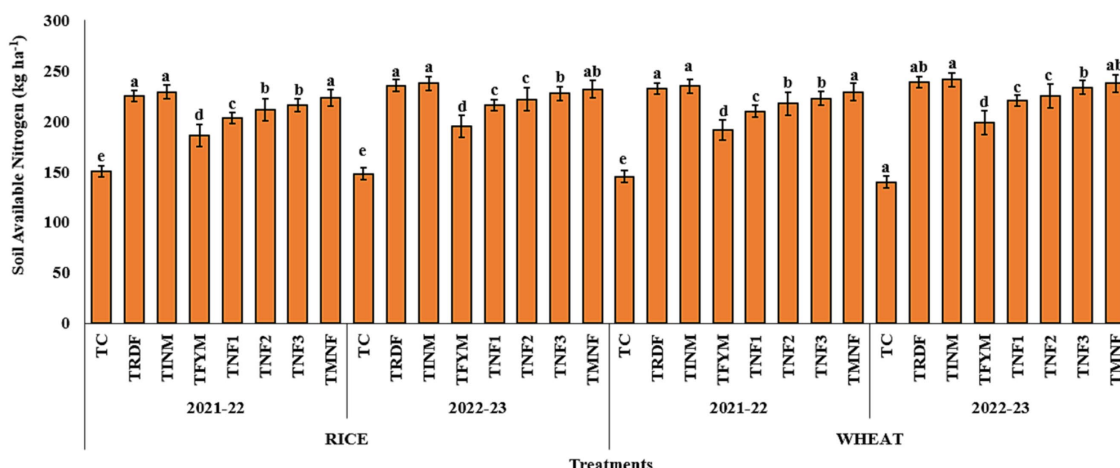


FIGURE 3 Effect of different nutrient management practices on soil available nitrogen. T_C: Control; T_{RDf}: Recommended dosage of fertilizer; T_{INM}: Integrated nutrient management; T_{FYM}: Farm yard manure; T_{NF-1}: Natural Farming-1; T_{NF-2}: Natural Farming-2; T_{NF-3}: Natural Farming-3; T_{MNF}: Modified Natural Farming, Brief details of the treatment is given in Table 1. Data refer to mean of three replicate and error bars depicts standard deviation. Bars with the same alphabet are not significantly different at $p \leq 0.05$. Different alphabet indicates statistically significant differences within each treatment at $p < 0.05$ (Duncan test).

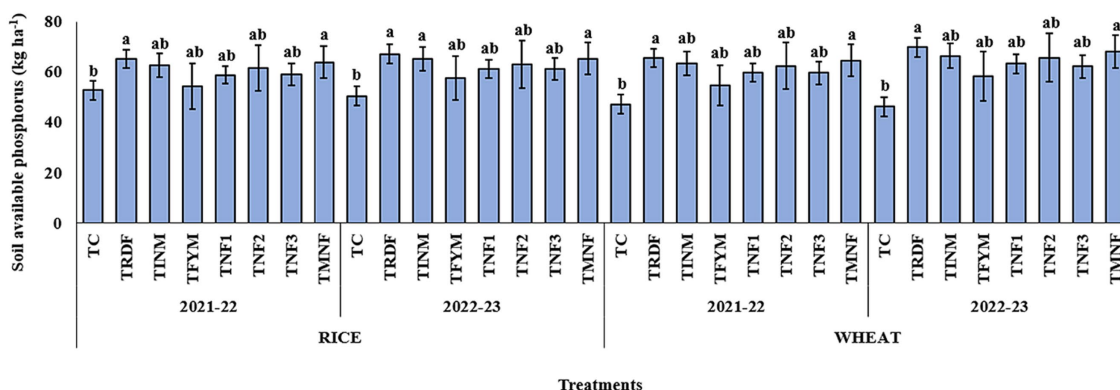


FIGURE 4 Effect of different nutrient management practices on soil available phosphorus. T_C: Control; T_{RDf}: Recommended dosage of fertilizer; T_{INM}: Integrated nutrient management; T_{FYM}: Farm yard manure; T_{NF-1}: Natural Farming-1; T_{NF-2}: Natural Farming-2; T_{NF-3}: Natural Farming-3; T_{MNF}: Modified Natural Farming, Brief details of the treatment is given in Table 1. Data refer to mean of three replicate and error bars depicts standard deviation. Bars with the same alphabet are not significantly different at $p \leq 0.05$. Different alphabet indicates statistically significant differences within each treatment at $p < 0.05$ (Duncan test).

control treatments consistently exhibited the lowest nitrogen uptake during both years.

3.5 Soil biological properties

3.5.1 Soil microbial biomass carbon and nitrogen

Our experimental result revealed that treatment receiving natural farming inputs exert a substantial influence on the Microbial biomass carbon (MBC) and Microbial biomass nitrogen (MBN) content in soil. In the present investigation, specifically, the treatment denoted as T_{NF1}, T_{NF2}, T_{NF3}, and T_{MNF} exhibited a notably pronounced impact on the soil microbial biomass, as indicated in (Table 5). In both years under study, 2021–22 and 2022–23, the research trial yielded highest MBC values in the T_{MNF} treatment for both rice (190.3 mg kg⁻¹) and wheat

(204.5 mg kg⁻¹) during the last year of experiment. T_{NF3} also exhibited a relatively higher MBC content, which was statistically equivalent to that of T_{MNF}. In the context of rice cultivation, T_{MNF} demonstrated a substantial 44.9% increase in MBC when compared to the T_{RDf} treatment. Similarly, during wheat cultivation, T_{MNF} exhibited a substantial 53.5% increase in MBC relative to the T_{RDf} treatment. Notably, analogous trends were observed for MBN content in the soil. The highest and lowest values for MBN were consistently recorded in the T_{MNF} and T_C treatments, respectively. During rice cultivation, T_{MNF} displayed an MBN content of 88.9 mg kg⁻¹ while T_C had a content of 20.4 mg kg⁻¹. During wheat cultivation, T_{MNF} exhibited an MBN content of 95.4 mg kg⁻¹, whereas T_C displayed a content of 21.8 mg kg⁻¹. This implies a remarkable percentage increase of 40.6% in MBN content during rice cultivation and an even more substantial 102.1% increase during wheat cultivation in the T_{MNF} treatment

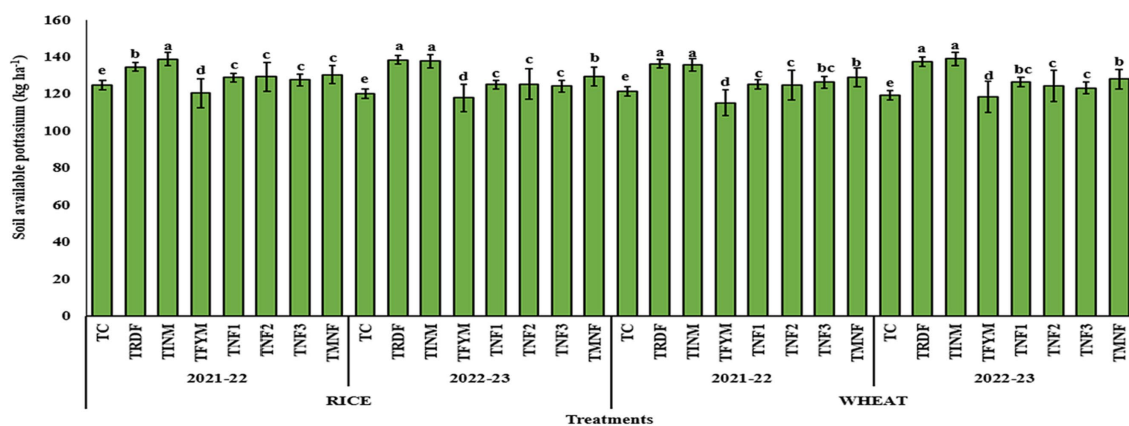


FIGURE 5
Effect of different nutrient management practices on soil available potassium. T_C, Control; T_{TRDF}, Recommended dosage of fertilizer; T_{TINM}, Integrated nutrient management; T_{IFYM}, Farm yard manure; T_{TNF-1}, Natural Farming-1; T_{TNF-2}, Natural Farming-2; T_{TNF-3}, Natural Farming-3; T_{TMNF}, Modified Natural Farming, Brief details of the treatment is given in Table 1. Data refer to mean of three replicate and error bars depicts standard deviation. Bars with the same alphabet are not significantly different at $p \leq 0.05$. Different alphabet indicates statistically significant differences within each treatment at $p < 0.05$ (Duncan test).

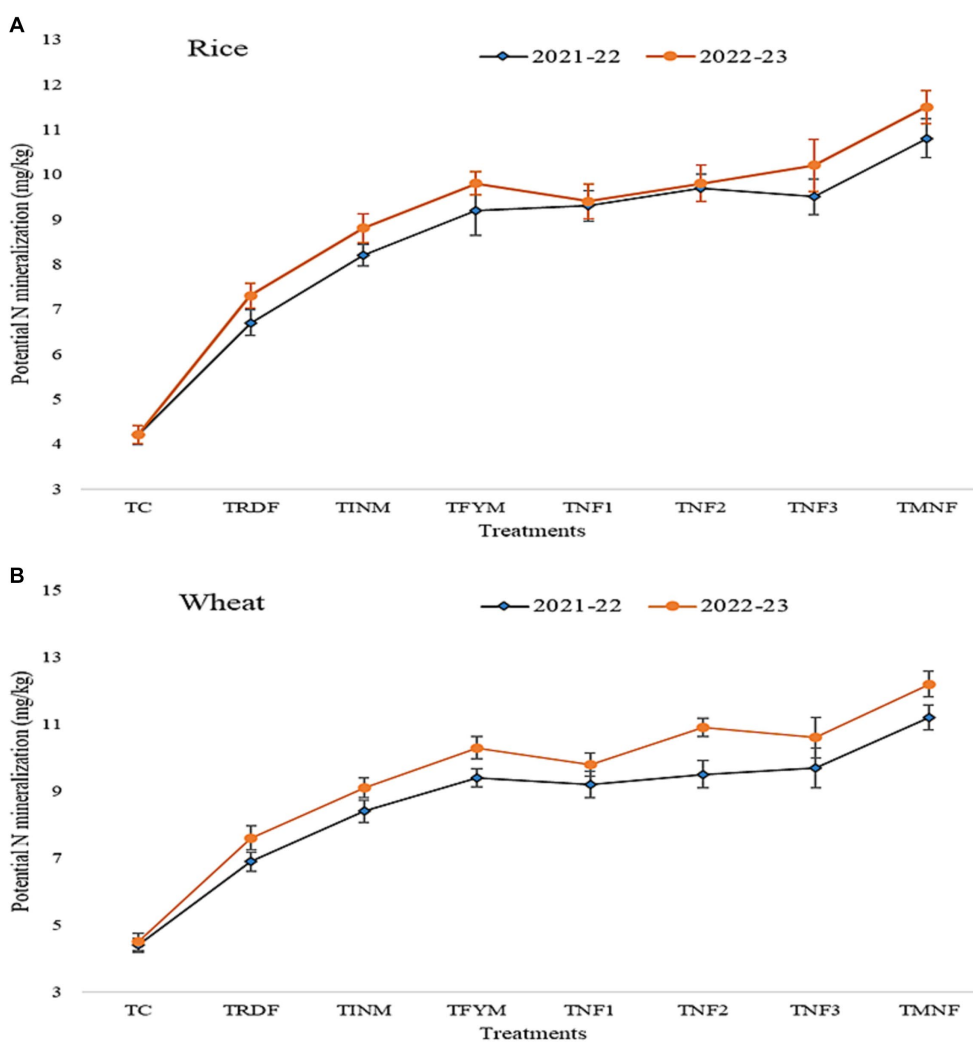


FIGURE 6
(A) and (B) explain as the effect different nutrient management practices on nitrogen mineralization for crop rice and wheat respectively. A is for Rice crop of 2021-22 and 2022-23 and B is for Wheat crop of 2021-22 and 2022-23.

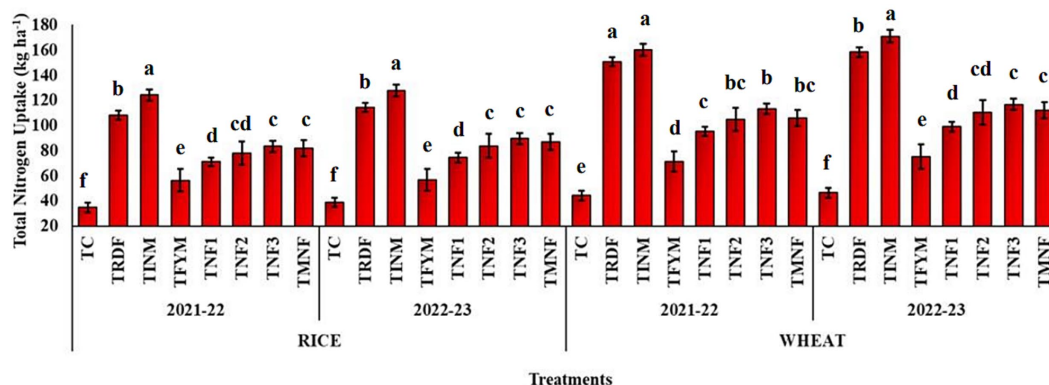


FIGURE 7 Effect of different nutrient management practices on total nitrogen uptake by rice and wheat during cropping season 2021–22 and 2022–23. [T_C, Control; T_{RDF}, Recommended dosage of fertilizer; T_{INM}, Integrated nutrient management; T_{FYM}, Farm yard manure; T_{NF-1}, Natural Farming-1; T_{NF-2}, Natural Farming-2; T_{NF-3}, Natural Farming-3; T_{MNF}, Modified Natural Farming. Brief details of the treatment is given in Table 1]. Data refer to mean of three replicate and error bars depicts standard deviation.

TABLE 5 Effect of different natural farming inputs (Ghanjeevamrit, Jeevamrit, and Beejamrit), organic (FYM) and in-organic fertilizer on soil MBC and MBN.

Treatment	Rice (2021–22)		Rice (2022–23)		Wheat (2021–22)		Wheat (2022–23)	
	MBC (mg/kg)	MBN (mg/kg)	MBC (mg/kg)	MBN (mg/kg)	MBC (mg/kg)	MBN (mg/kg)	MBC (mg/kg)	MBN (mg/kg)
T _C	75.2 ^g	18.2 ^g	79.4 ^f	20.4 ^g	78.3 ^b	19.4 ^g	81.5 ^f	21.8 ^g
T _{RDF}	125.4 ^e	39.5 ^f	131.3 ^c	45.3 ^f	127.5 ^g	42.5 ^f	133.8 ^e	47.2 ^f
T _{INM}	136.9 ^d	45.8 ^e	145.2 ^d	57.9 ^e	139.7 ^c	52.6 ^e	152.3 ^d	62.3 ^e
T _{FYM}	131.4 ^e	51.4 ^d	142.4 ^d	60.3 ^d	135.3 ^f	55.2 ^d	162.4 ^c	65.4 ^d
T _{NF1}	147.7 ^c	64.4 ^c	165.3 ^c	79.5 ^c	154.5 ^d	68.3 ^c	179.8 ^b	85.3 ^c
T _{NF2}	149.2 ^c	68.9 ^a	179.5 ^b	81.5 ^b	158.7 ^c	72.2 ^a	183.6 ^b	86.4 ^c
T _{NF3}	158.4 ^a	66.4 ^c	188.2 ^a	80.4 ^{bc}	166.6 ^a	70.7 ^c	201.3 ^a	89.2 ^b
T _{MNF}	154.8 ^b	63.6 ^b	190.3 ^a	88.9 ^a	162.7 ^b	67.2 ^b	204.5 ^a	95.4 ^a
Sem	1.25	0.43	1.35	0.61	1.25	0.52	1.45	0.66
LSD (p=0.05)	3.79	1.31	4.10	1.84	3.79	1.58	4.39	1.99

T_C, Control; T_{RDF}, Recommended dosage of fertilizer; T_{INM}, Integrated nutrient management; T_{FYM}, Farm yard manure; T_{NF-1}, Natural Farming-1; T_{NF-2}, Natural Farming-2; T_{NF-3}, Natural Farming-3; T_{MNF}, Modified Natural Farming. Brief details of the treatment is given in Table 1. Means followed by the same letter(s) with in column do not differ significantly at 5% level probability level by DMRT.

compared to the TRDF treatment. Conversely, the lowest MBC levels were observed in the T_C treatment, registering at 79.4 mg kg⁻¹ and 78 mg kg⁻¹ following the harvest of two consecutive rice and wheat seasons, respectively.

3.5.2 Soil enzymatic activity index

An important innovation in this study was the introduction of the integrated index GMenz, which serves as a dimensionless parameter facilitating the comparison of combined enzyme activity (dehydrogenase, β-glucosidase and urease) and soil sample quality. The GMenz values ranged from 7.85 to 32.83 across all treatments for both rice and wheat during a two-year experiment as shown in Figure 8. Over the course of this research, T_{MNF} consistently exhibited the highest GMenz value, regardless of the crop type and year. At the end of the 2022–23 wheat crop season, T_{MNF} displayed a remarkable 77.03% increase in GMenz value compared to T_{RDF}, and a substantial 48.5% increase compared to T_{INM}. In contrast, TC registered the

lowest GMenz value, which was 7.99, making it four times lower than T_{MNF}.

3.5.3 Soil microbial population

Throughout the research conducted in both the years 2021–22 and 2022–23, the investigation revealed that the highest bacterial population was consistently observed in the T_{MNF} treatment, with subsequent populations in T_{NF3}, indicating (33.8 and 34.4 × 10⁶ cfu g⁻¹ soil) during rice-wheat crop in the respective years. Conversely, the T_{RDF} treatment exhibited significantly lower bacterial populations, registering only (19.4 and 20.2 × 10⁶ cfu g⁻¹ soil) compared to all other treatments utilizing natural farming inputs. This data underscores a substantial 70.2% increase in bacterial population in T_{MNF} as compared to the T_{RDF} treatment during the final crop. It further elucidates the findings related to the fungal and actinomycetes populations in the soil for the years 2021–22 and 2022–23. Remarkably, the T_{MNF} treatment consistently displayed the highest populations, recording

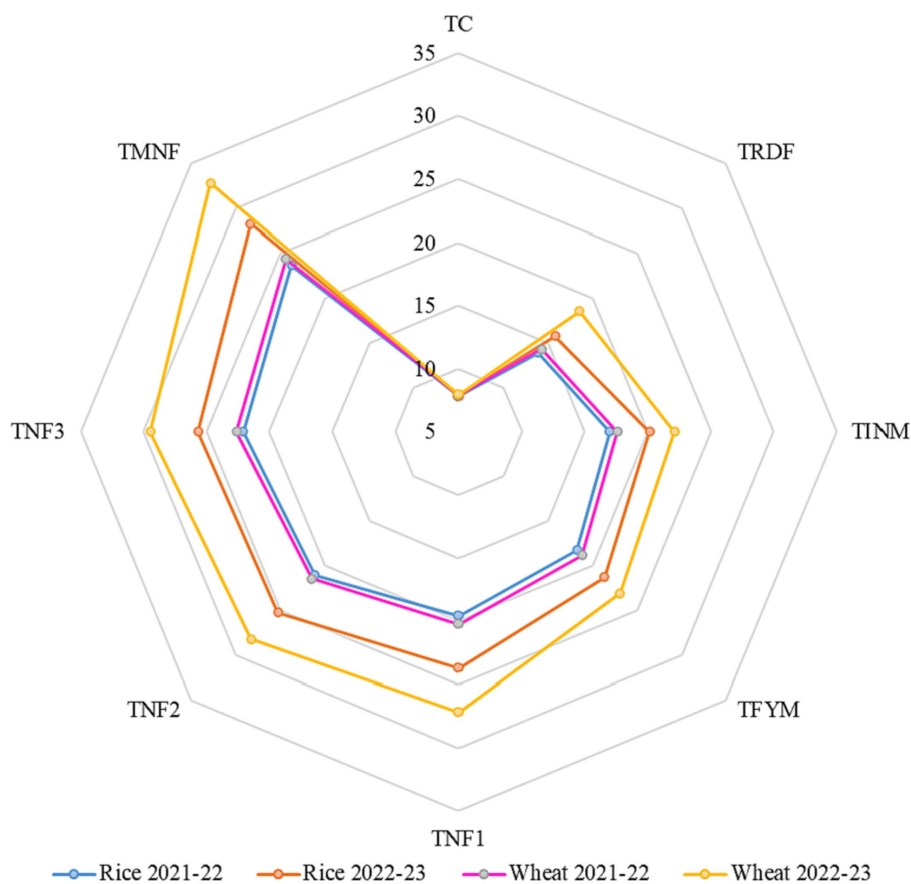


FIGURE 8

Effect of different nutrient management practices on the geometric mean of assayed enzyme activities (relative value). [T_C, Control; T_{RDF}, Recommended dosage of fertilizer, T_{INM}, Integrated nutrient management; T_{FYM}, Farm yard manure; T_{NF-1}, Natural Farming-1; T_{NF-2}, Natural Farming-2; T_{NF-3}, Natural Farming-3; T_{MNF}, Modified Natural Farming, Brief details of the treatment is given in Table 1].

10.3 and $11.5 \times 10^3 \text{ cfu g}^{-1}$ soil for fungi and 23.5 and $24.8 \times 10^5 \text{ cfu g}^{-1}$ soil for actinomycetes in the respective years. Following this, T_{NF3} exhibited the second-highest fungal population with 9.8 and $10.4 \times 10^3 \text{ cfu g}^{-1}$ soil, whereas T_{NF2} recorded the second-highest actinomycetes population with 23.6 and $24.8 \times 10^5 \text{ cfu g}^{-1}$ soil. Conversely, the control treatment consistently demonstrated the lowest fungal and actinomycetes populations, measuring only (4.1 and $4.3 \times 10^3 \text{ cfu g}^{-1}$ of soil for fungi and 11.4 and $11.7 \times 10^5 \text{ cfu g}^{-1}$ of soil) for actinomycetes during the respective years. These observations indicate a consistent increase in microbial counts over time. Importantly, when the organic inputs, including Farm Yard Manure (FYM), Ghanjeevamrit, Jeevamrit, and mulching, were applied collectively, the microbial population in the soil exhibited a much higher count than when any of these inputs were applied individually (Figures 9, 10).

3.6 Crop yield

The outcome of a crop, which is often contingent upon the development of yield attributes, is primarily determined by the grain yield. The utilization of an integrated nutrient source substantially enhances the grain yield of rice and wheat. Particularly, during the end of the Kharif season in 2022, the highest grain yield, amounting to

4.76 t ha^{-1} , was observed in the treatment denoted as T_{INM} when compared to the T_{RDF} treatment and other treatments employing natural farming inputs such as T_{NF1}, T_{NF2}, T_{NF3}, and T_{MNF}. Notably, T_{INM} demonstrated a remarkable 9.3% increase in rice grain yield in comparison to T_{RDF}, which yielded 4.31 t ha^{-1} . Among the various natural farming inputs, T_{MNF} yielded the highest grain yield at 4.08 t ha^{-1} , although this result was not statistically distinguishable from the yield of T_{NF2}, which amounted to 3.81 t ha^{-1} . Over the course of the two-year experimental trial, spanning from 2021–22 to 2022–23, it was evident that significant differences in wheat yield existed among the various treatments. The highest wheat yield recorded at the end of the experimental trial was in the T_{RDF} treatment, yielding 5.82 t ha^{-1} , which was statistically equivalent to the yield of 5.70 t ha^{-1} observed in the T_{INM} treatment. There was a notable decrease in yield observed in the treatments employing natural farming inputs. Among the natural farming inputs, T_{NF2} yielded the highest wheat yield at 4.19 t ha^{-1} , which was not statistically distinct from the yield of 4.14 t ha^{-1} observed in the T_{MNF} treatment (Figure 11).

3.7 Correlation analysis

Correlation analysis of (BP: bacterial population; FP: fungal population; AP: actinomycetes population; MBC: microbial biomass

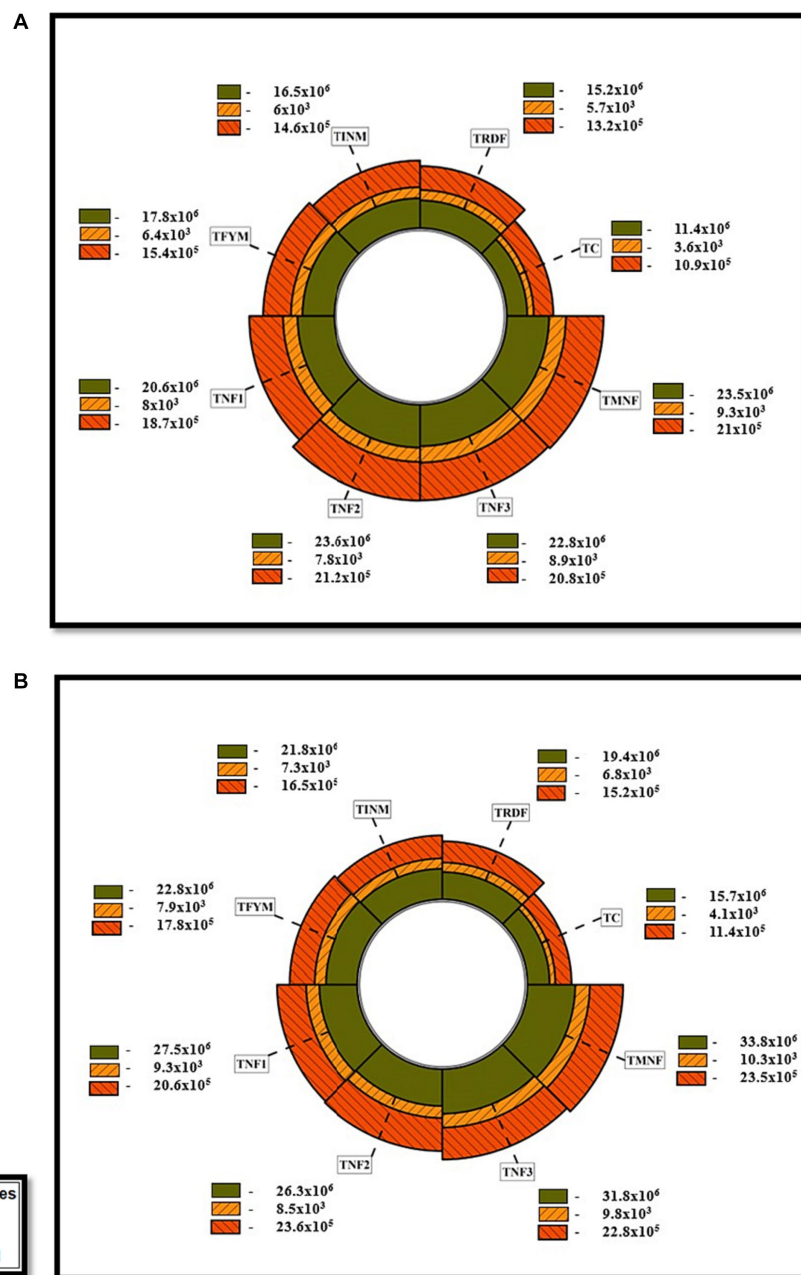


FIGURE 9 Effect of different natural farming inputs (Ghanjeevramrit, Jeevamrit, and Beejamrit), organic (FYM) and in-organic fertilizer on soil microbial population (A) rice crop (2021–22) and (B) rice crop (2022–23). LSD value ($p > 0.05$): for Bacteria 0.70×10^6 and 0.67×10^6 , Fungi 0.17×10^3 and 0.22×10^3 and Actinomycetes 0.43×10^5 and 0.50×10^5 in 2021–22 and 2022–23, respectively. [T_C, Control; T_{RDF}, Recommended dosage of fertilizer; T_{TINM}, Integrated nutrient management; T_{TFYM}, Farm yard manure; T_{TNF-1}, Natural Farming-1; T_{TNF-2}, Natural Farming-2; T_{TNF-3}, Natural Farming-3; T_{TMNF}, Modified Natural Farming, Brief details of the treatment is given in Table 1]. Data refer to mean of three replicates as shown in radial bar on a polar coordinate system.

carbon; MBN: microbial biomass nitrogen; DHA: dehydrogenase activity; UrA: urease activity; BGA: beta-glucosidase activity; PMN: potential mineralizable nitrogen) for rice and wheat crop during 2021–22 and 2022–23 has been shown in Figures 12, 13. The PMN during flowering stage was significantly ($p = 0.001$) positive correlated with BP ($R^2 = 0.818$ and 0.901), FP ($R^2 = 0.929$ and 0.942), AP ($R^2 = 0.890$ and 0.906), MBC ($R^2 = 0.918$ and 0.942), MBN ($R^2 = 0.926$ and 0.940), DHA ($R^2 = 0.834$ and 0.801), UrA ($R^2 = 0.748$ and 0.742) and BGA ($R^2 = 0.972$ and 0.973) in rice and wheat, respectively.

4 Discussion

The study was conducted to evaluate the effect of different natural farming and organic inputs on nitrogen uptake, soil biological health and crop yield in rice-wheat cropping system. After the rice and wheat harvests spanning from 2021 to 2023, it was observed that the soil's pH and electrical conductivity (EC) remained relatively stable, despite the application of diverse natural farming inputs. Notably, a minor reduction in pH was observed in the T_{RDF}, while all other treatment

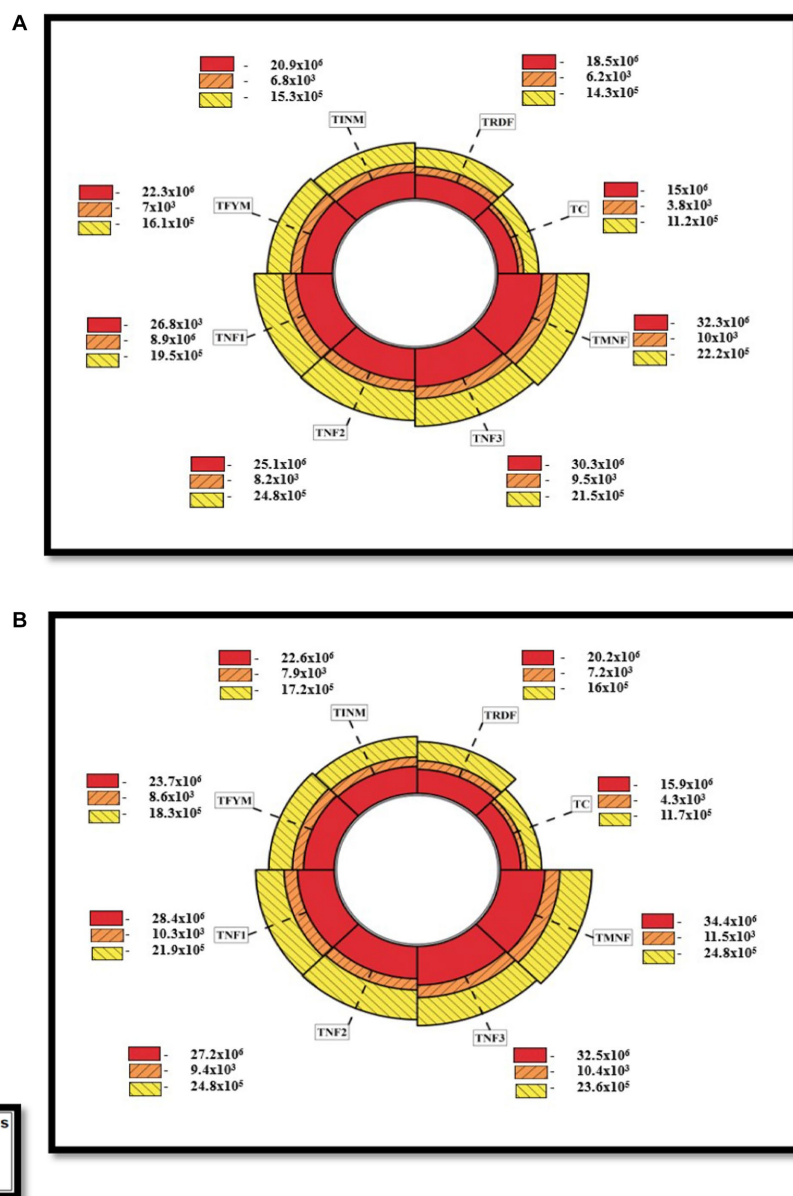


FIGURE 10 Effect of different natural farming inputs (Ghanjeevamrit, Jeevamrit, and Beejamrit), organic (FYM) and in-organic fertilizer on soil microbial population during (A) wheat crop (2021–22) and (B) wheat crop (2022–23). LSD value ($p > 0.05$): for Bacteria 0.74×10^6 and 0.69×10^6 , Fungi 0.23×10^3 , and 0.24×10^3 and Actinomycetes 0.52×10^5 and 0.53×10^5 in 2021–22 and 2022–23, respectively. [T_C, Control; T_{RDF}, Recommended dosage of fertilizer; T_{TINM}, Integrated nutrient management; T_{TFYM}, Farm yard manure; T_{TNF-1}, Natural Farming-1; T_{TNF-2}, Natural Farming-2; T_{TNF-3}, Natural Farming-3; T_{TMNF}, Modified Natural Farming. Brief details of the treatment is given in Table 1]. Data refer to mean of three replicates as shown in a radial bar chart on a polar coordinate system.

groups showed slight increases in pH and EC values over time (Yadav et al., 2022). Soil organic matter by interacting with both biotic and abiotic soil component plays a significant role in improving soil health and restoring soil fertility (Gurmu, 2019). Numerous studies have investigated the comparative impacts of synthetic fertilizers and organic amendments on soil properties and crop production. It was found that the enhanced organic carbon content in the soil, resulting from the application of cow-based solid and liquid manure, could be attributed to the direct incorporation of organic materials and improved root growth. Subsequent decomposition of these organic

materials appeared to contribute to the increased organic carbon content in the soil (Choudhary et al., 2022; Rautela et al., 2022). In our own investigation, the higher organic carbon content in all soil samples treated with natural farming inputs and organic amendments was primarily associated with the introduction of organic materials in comparison to mineral fertilizer and untreated control groups (Hu et al., 2023). Soil nutrient availability is a dynamic process that is highly responsive to intensive agricultural practices, environmental changes, and is typically constrained by both the quantity and quality of nutrients supplied through various physiochemical mechanisms

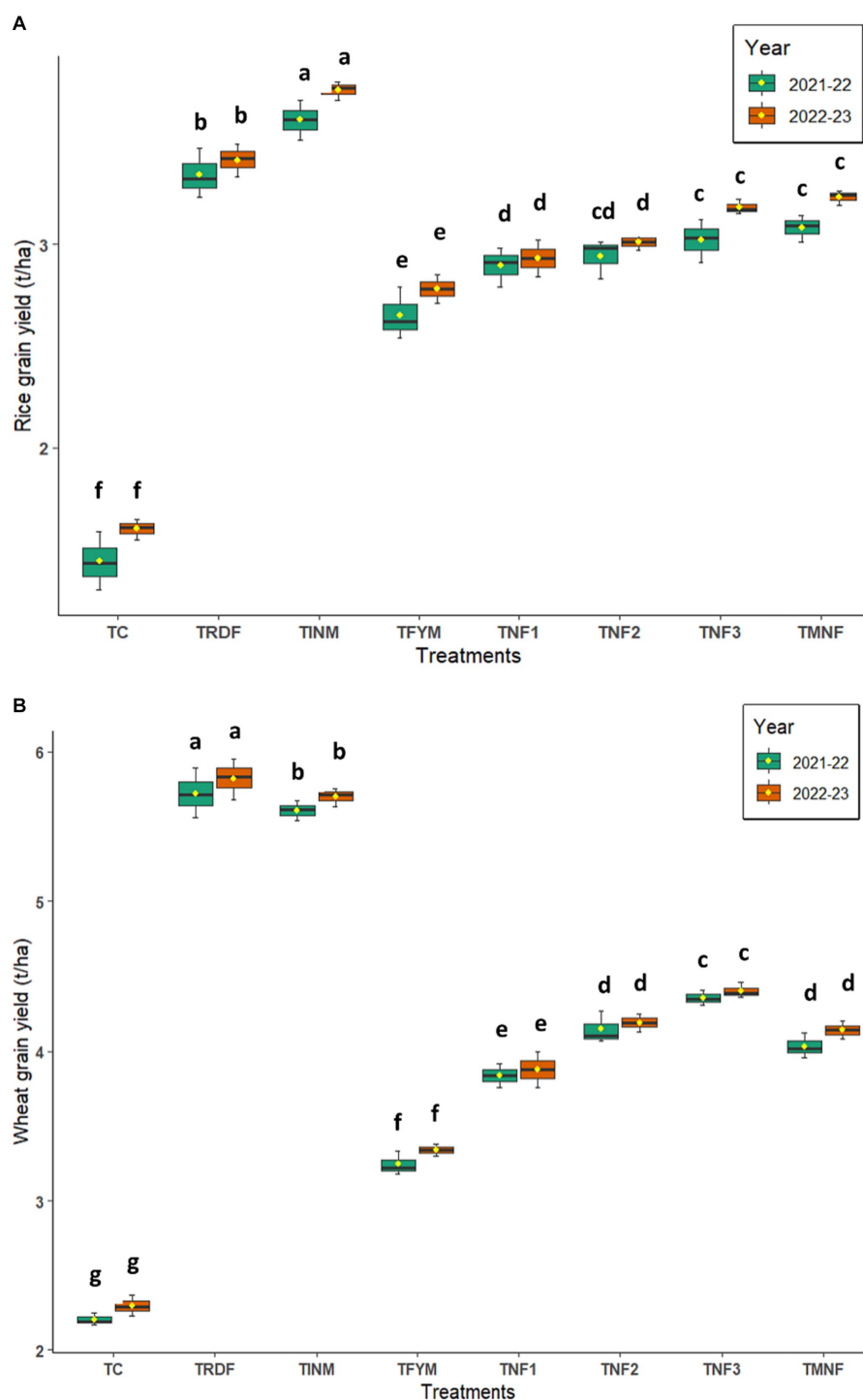


FIGURE 11 Box plot of different natural farming inputs (Ghanjeevamrit, Jeevamrit, and Beejamrit), organic (FYM) and in-organic fertilizers on (A) rice grain yield and (B) wheat grain yield. [TC, Control; TRDF, Recommended dosage of fertilizer; TINM, Integrated nutrient management; TFYM, Farm yard manure; TNF-1, Natural Farming-1; TNF-2, Natural Farming-2; TNF-3, Natural Farming-3; TMNF, Modified Natural Farming, Brief details of the treatment is given in Table 1]. Box plots of chemicals analyzed in the present study (logarithmic scale).

(Rana et al., 2021). Previous research has consistently demonstrated a positive correlation between the use of various organic amendments and increased nutrient availability in soil (Darjee et al., 2023; Ramalingappa et al., 2023). In our current investigation, the incorporation of Ghanjeevamrit, Jeevamrit, Beejamrit, and Farm Yard

Manure (FYM) resulted in a notable enhancement of the pool of available major nutrients in the soil. This phenomenon could be attributed to the accelerated mineralization of nitrogen, facilitated by heightened microbial activity within these treatments, in conjunction with the application of natural farming inputs (Darjee

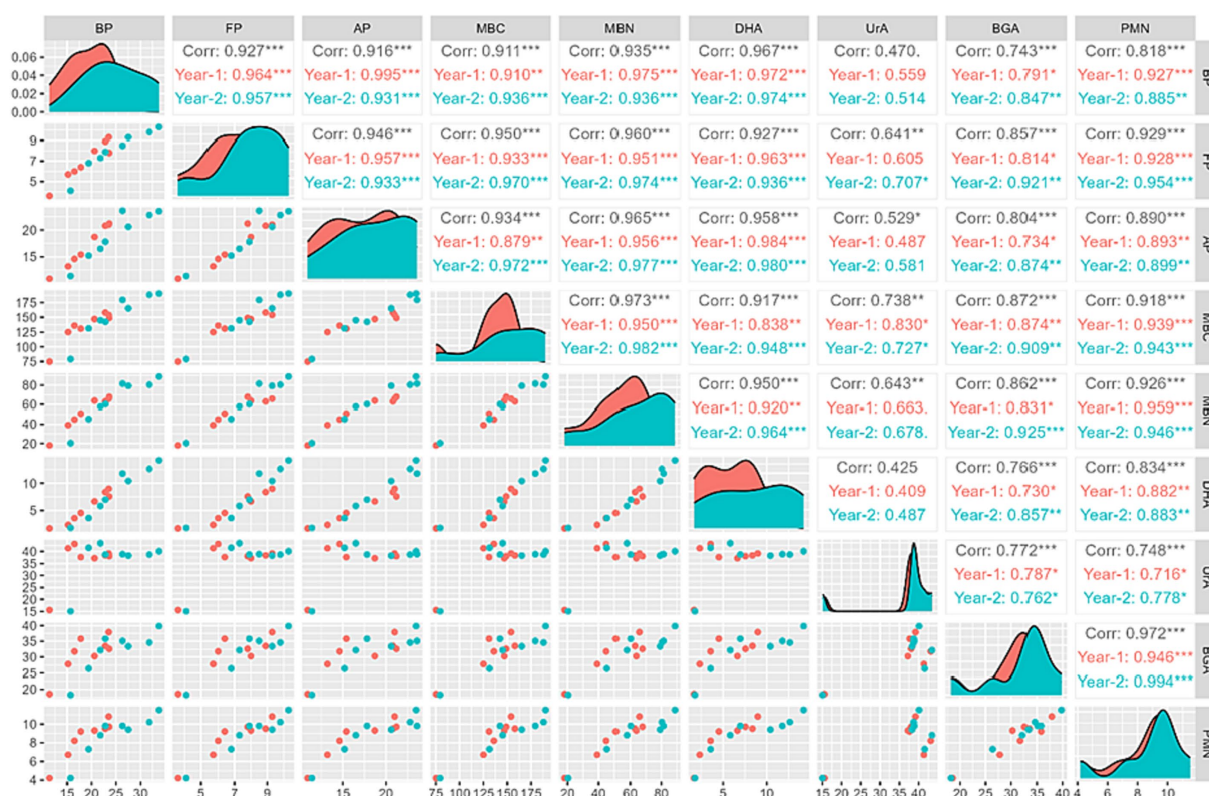


FIGURE 12

Correlation panel graph in rice (BP, bacterial population; FP, fungal population; AP, actinomycetes population; MBC, microbial biomass carbon; MBN, microbial biomass nitrogen; DHA, dehydrogenase activity; UrA, urease activity; BGA, beta-glucosidase activity; PMN, potential mineralizable nitrogen).

et al., 2022). Aulakh et al. (2018) also reported similar findings while studying the impact of farmyard manure and Jeevamrit in a maize-wheat organic production system in Punjab. However, some studies have indicated an increase in available nitrogen in the soil when optimizing Jeevamrit doses and application timing to boost wheat productivity under natural farming systems (Kaur and Saini, 2021). During our study, variations in the available phosphorus content were observed among the different treatment groups. This divergence may be due to the increased release of organic acids during mineralization in the case of treatments combining Ghanjeevamrit and Jeevamrit, which enhanced soil phosphorus levels by rendering native phosphates more soluble (Naresh et al., 2018). Interestingly, similar observations of increased soil available phosphorus content were made in studies focused on the application of Jeevamrit to enhance soil properties in zero-budget natural farming fields (Saharan et al., 2023). Our findings align with those of Sharma and Chadak (2022), who investigated residual soil fertility, nutrient uptake, and okra yield influenced by bioorganic nutrient sources. Notably, treatments with organic amendments exhibited significantly lower values of soil available potassium, possibly due to the absence of external potassium inputs and the depletion of the local potassium reservoir by plants. This depletion, however, was mitigated by the accumulation of microflora and fauna resulting from the addition of Jeevamrit and Ghanjeevamrit (Al-Jabori et al., 2011). Over the course of our research trial, an increase in potassium content was observed, which may be attributed to the ability of various natural farming amendments to increase soil

potassium availability, promote potassium release, and reduce potassium fixation within the soil (Bader et al., 2021). This aligns with the findings of Wafaa and Mona (2015), who noted that the decomposition of organic fertilizers facilitates the dissolution of potassium-containing minerals in response to naturally occurring acids, such as fulvic and humic acids. This process releases potassium ions, subsequently elevating their concentration in the soil solution. The pattern of nitrogen uptake exhibited an elevated trend in both conventional fertilization methods and integrated nutrient management practices. Specifically, the microbial inoculum, or Jeevamrit, applied at various intervals to the soil, played a pivotal role in augmenting nutrient availability and subsequently enhancing enzymatic activity. As noted by Choudhary et al. (2022), Jeevamrit as well as Ghanjeevamrit significantly amplifies microbial activity in the soil, thereby enhancing the accessibility of nutrients to crops. Nitrogen availability frequently presents limitations within agro-ecosystems. Soil organic matter stands as the primary source of nitrogen for both crops and microorganisms, achieved through the process of mineralization. Superior-quality organic matter is characterized by a low carbon-to-nitrogen (C: N) ratio and an ample nitrogen content, sufficient to support the growth of microorganisms and crops. In most scenarios, soil suffers from nitrogen deficiency, primarily due to its meager or substandard organic matter.

The introduction of natural farming inputs, which tend to be nitrogen-rich, to the soil typically enhances the quality of soil organic matter. Nitrogen mineralization, on the other hand, exhibits

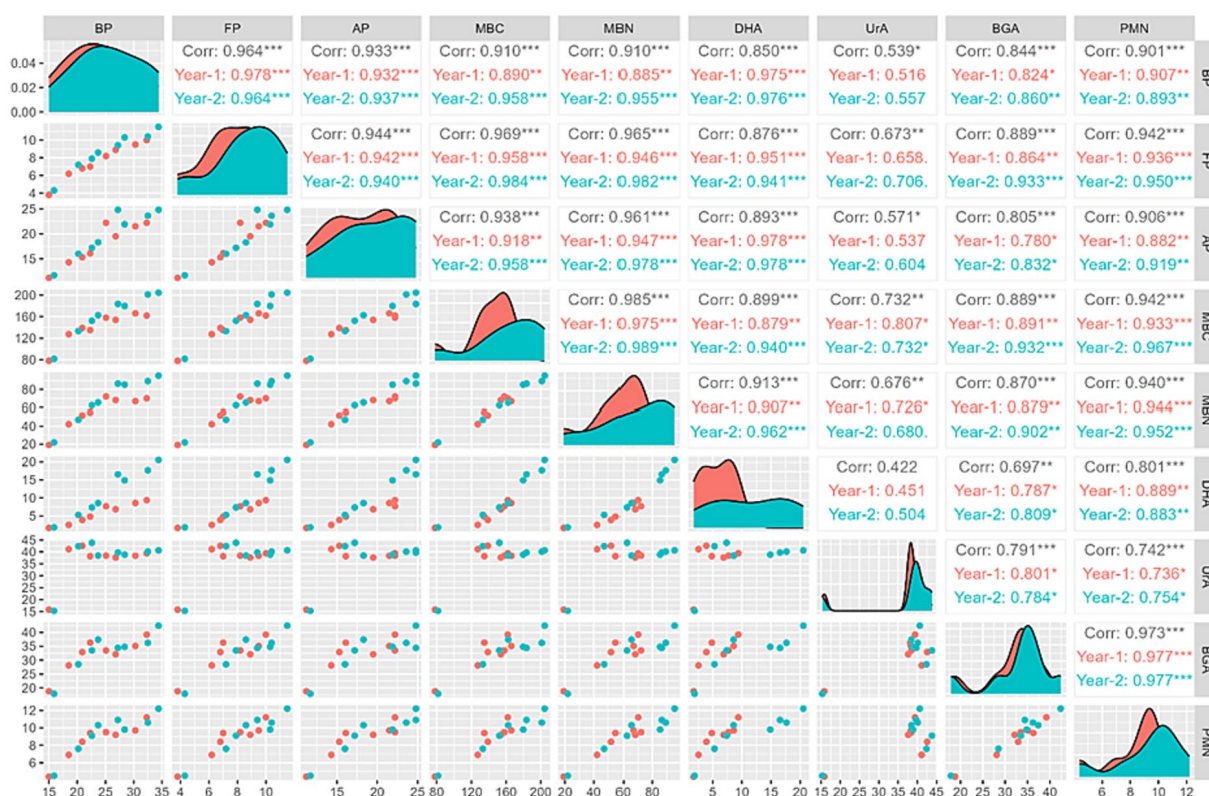


FIGURE 13

Correlation panel graph in wheat (BP, bacterial population; FP, fungal population; AP, actinomycetes population; MBC, microbial biomass carbon; MBN, microbial biomass nitrogen; DHA, dehydrogenase activity; UrA, urease activity; BGA, beta-glucosidase activity; PMN, potential mineralizable nitrogen).

variances among the various natural farming and organic inputs, with the most pronounced nitrogen mineralization observed in the treatment receiving an integrated blend of Farm Yard Manure (FYM), Ghanjeevamrit, and Jeevamrit (citations). The composting process serves to stabilize organic compounds, diminishing the proportion of soluble carbon and nitrogen forms (Masunga et al., 2016). In another study, it was revealed that only a minor fraction of organic nitrogen in composts underwent mineralization over time (Zhao et al., 2013). Consequently, even when manure is abundant in nitrogen, the release of nitrogen into the soil proceeds at a slow pace. Tits et al. (2014) noted restricted nitrogen release in composts derived from vegetable, fruit, and garden waste, despite their elevated nitrogen content (1.5% of fresh weight). Lazicki et al. (2020) also observed that nitrogen mineralization was more substantial in animal manure, such as cow dung, compared to composts (7 and 1% of applied organic nitrogen, respectively). This was the case despite their similar C:N ratios and nitrogen contents (10.1 and 9.3, respectively, and 1.2 and 2.2%, respectively).

In the current investigation, it has been determined that soil microbial carbon and nitrogen are profoundly influenced by both organic and natural farming inputs, namely Ghanjeevamrit, Jeevamrit, and Beejamrit. This influence may be attributed to the substantial presence of organic matter in the soil, which provides a nutrient-rich substrate conducive to microbial proliferation (Hicks et al., 2021). The improved MBC and MBN compare in natural farming inputs plot might be due to residue retention (mulching), tillage reduction as well

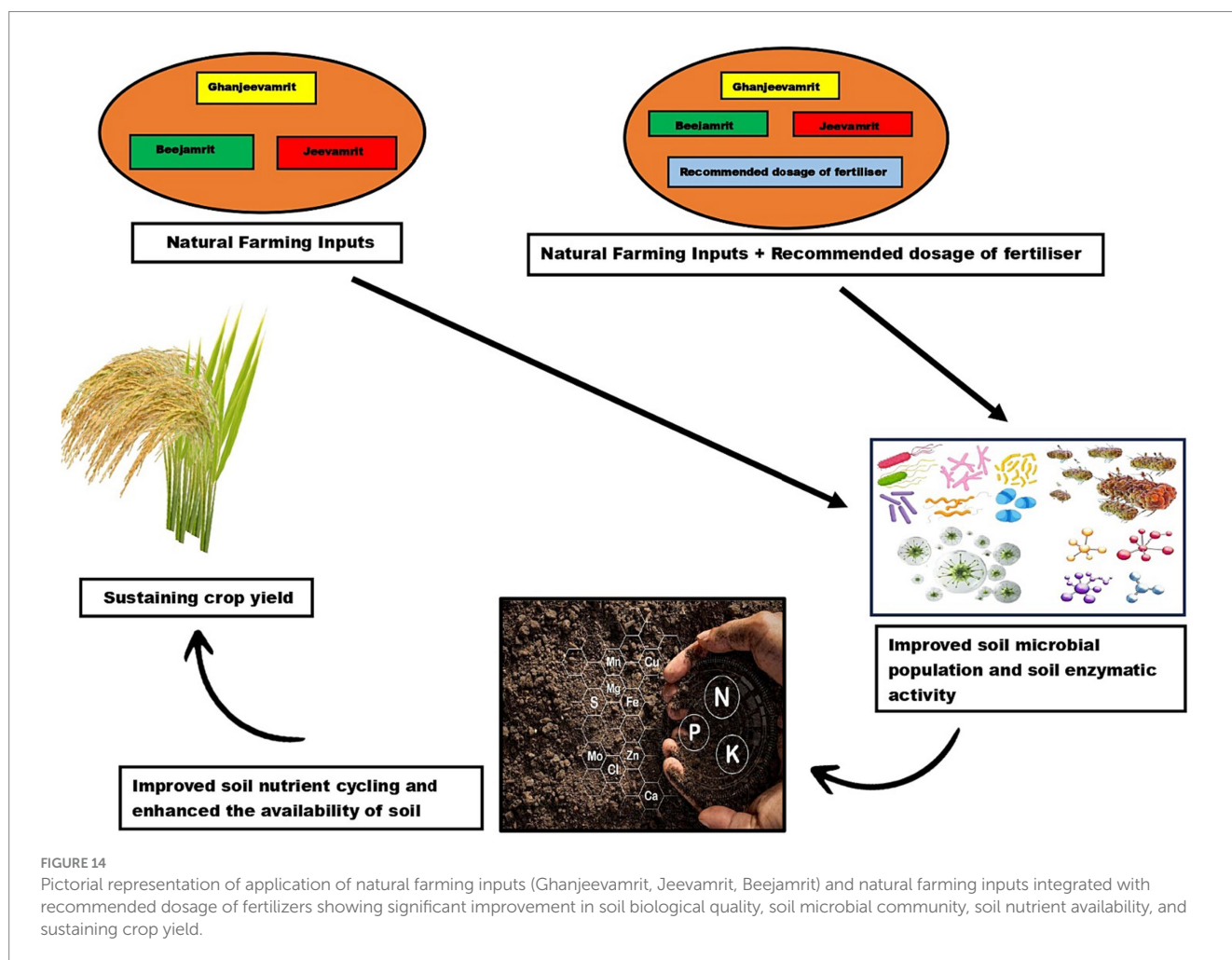
as addition of Farm Yard Manure (FYM), liquid manure (Jeevamrit), and solid manure (Ghanjeevamrit), are incorporated into the soil, they serve as a nutritional source for microorganisms as well as, supply of organic matter provided sufficient C resources for the growth and reproduction of soil microorganisms which in turn resulting in an accumulation of microbial biomass carbon and nitrogen, whereas absence of residue retention (mulching) and organic manure and presence of conventional tillage in control plot would have resulted in such low MBC and MBN content (Ramalingappa et al., 2023). Microbial Biomass Carbon (MBC) is notably more responsive to cultural practices than Soil Organic Carbon (SOC) and represents a substantial component of SOC. In soils treated with natural farming inputs, the highest levels of MBC and Microbial Biomass Nitrogen (MBN) were recorded (Yang et al., 2018). This observation suggests that natural farming inputs may have functioned as substrates and sources of soluble nutrients conducive to the proliferation of soil microorganisms. These findings align with previous research, indicating that the application of Jeevamrit substantially elevates soil MBC and MBN (Rathore et al., 2023). Soil enzymatic activity is frequently employed as a measure of soil quality, relates to soil nutrient transformation and also strongly correlated with the amount of organic matter in the soil (Merino et al., 2019). The enzymes selected in the current study, are dehydrogenase, β -glucosidase and urease, which are one of the most adequate, important and most sensitive bioindicators, relating to soil fertility (Wolińska and Stępniewska, 2012). Furthermore, according to Choudhary et al. (2022), there was

a favourable correlation between soil enzymatic activity and microbial biomass. The utilization of organic sources in the soil may enhance dehydrogenase activity as well as β -glucosidase by increasing substrate availability, promoting biological activity, and stabilizing extracellular enzymes through humic substance complexation (Adak et al., 2014). Higher enzyme activity was seen in the organic amendment treatments, such as T_{INM} , T_{FYM} , T_{NF1} , T_{NF2} , T_{NF3} , and T_{MNF} which is consistent with the results reported by Rao et al. (2019), Ghasal et al. (2023) and suggests that these treatments can enhance the cycling of soil nutrients (such as C, N, and P). Soil enzymatic activity is a commonly utilized metric for assessing soil quality, as it is intricately linked to soil nutrient transformations and exhibits a robust correlation with the quantity of organic matter within the soil (Merino et al., 2019). In the present study, the selected enzymes, specifically dehydrogenase, β -glucosidase, and urease, stand as highly suitable, crucial, and exceedingly sensitive bioindicators with strong ties to soil fertility (Wolińska and Stępniewska, 2012). Moreover, in accordance with Choudhary et al. (2022), a positive association was observed between soil enzymatic activity and microbial biomass. The incorporation of organic sources into the soil has the potential to elevate dehydrogenase and β -glucosidase activities by augmenting the availability of substrates, fostering biological activity, and stabilizing extracellular enzymes through complexation with humic substances (Adak et al., 2014). Notably, enhanced enzyme activity was evident in treatments involving organic amendments such as T_{INM} , T_{FYM} , T_{NF1} , T_{NF2} , T_{NF3} , and T_{MNF} . These findings align with results reported by Rao et al. (2019) and Ghasal et al. (2023), indicating that these treatments have the capacity to bolster the cycling of soil nutrients, including carbon (C), nitrogen (N), and phosphorus (P). Soil biodiversity is recognized as a fundamental component underpinning the integrity, functionality, and long-term sustainability of soil ecosystems (Zhao et al., 2014). In our investigation, it was observed that the microbial community, in terms of richness, exhibited relatively higher values in the treatment receiving Ghanjeevamrit, Jeevamrit, and Beejamrit, in comparison to other fertilizer regimens. However, it's worth noting that while these responses were statistically significant when compared to the inorganic fertilizer treatment and the control group, they did not reach statistical significance when compared among the treatments utilizing various natural farming inputs. These findings align with the outcomes of Patro et al. (2009), which emphasizes that the metabolic activity of the microbial population in the soil is reflected in soil enzyme activity. The improved microbial population in the soil may be attributed to the combined impact of Ghanjeevamrit, Jeevamrit, and mulching (Saharan et al., 2023). Moreover, Ghanjeevamrit was found to have positive effects on soil properties, including the potential reduction of bulk density (Kumari et al., 2022), enhancement of soil aeration, and the provision of carbon as an energy source for the multiplication and survival of microbes present in Jeevamrit (Dhiman et al., 2023). According to Siddappa (2015), the application of Jeevamrit alone may lead to noticeably lower microbial activity because fungi, bacteria, and actinomycetes lack access to an organic carbon source necessary for their further proliferation. Conversely, the application of Ghanjeevamrit alone may result in a deficiency of microbial inoculum, which is typically present in Jeevamrit. Such a treatment approach might contribute to a more stable agroecosystem and promote sustainable crop production. In the present study, variations in crop yields over time were evident in response to the diverse fertilizer regimens applied. The majority of characteristics

contributing to yield exhibited an increase, primarily attributed to improved growth characteristics resulting from heightened nitrogen absorption and the efficient translocation of assimilates from source to sink (Mangaraj et al., 2022). This upturn in grain yield can be linked to the appropriate allocation of resources from source to sink and the timely availability of nutrients (Laila et al., 2022). Aulakh et al. (2018), while conducting research on nutrient sources for organic rice (*Oryza sativa*)-wheat (*Triticum aestivum*) cropping systems in north-west India, reported similar outcomes. Darjee et al. (2022) and Iqbal et al. (2022) also documented enhanced growth attributes of rice and wheat yields in plots where Farm Yard Manure (FYM) was combined with chemical fertilizers. These results can likely be ascribed to the fulfillment of the crop's nutritional requirements through improved nutrient availability throughout the crop's life cycle, thereby enhancing growth and ultimately resulting in higher wheat seed yields. These findings closely align with those of Sutar et al. (2019), who observed similar outcomes with the application of Jeevamrit at a rate of 1,000 L/ha. Additionally, Gupta and Bhadauria (2022) reported an increase in wheat yields when investigating the impact of zero-budget natural farming on nutrient content and uptake in wheat (*Triticum aestivum* L.).

5 Conclusion

Our experimental trial concluded that application of natural farming inputs (Ghanjeevamrit, Jeevamrit, Beejamrit) in rice-wheat cropping system, had shown significant improvement in soil biological quality and soil microbial community as shown in (Figure 14). The data on the build-up of macro nutrient (N, P, K) also improved. Remarkably, a considerable boost in nitrogen uptake was observed in crops treated with natural farming inputs over control treatment. This phenomenon can be ascribed to the expedited mineralization of native and applied nutrients, facilitated by the accumulation of microflora. In contrast the treatment with exclusive use of in-organic fertilizer had an adverse impact on soil biological quality, reduced microbial richness and poor soil enzymatic activity. The natural farming inputs, employing Ghanjeevamrit, Jeevamrit, Beejamrit, and farmyard manure (FYM) in combination resulted in significant higher grain yield of rice and wheat than control and treatment having FYM only but lower yields than treatments supplied nutrients through integrated nutrient management and in-organic fertilizers. Overall, our research suggests that introducing natural farming inputs into intensive agricultural systems can mitigate the detrimental effects resulting from excessive use of synthetic fertilizers. However, to determine the optimal strategy, further investigations into the types, application rates, and frequency of these natural farming inputs are essential. These studies should also consider the interplay between various soil types and the specific cultivated crop species. Additionally, extensive research on the characterization of natural farming inputs are essential to substantiate our findings on a larger scale. Further investigations should focus on developing site-specific and comprehensive packages of practices to enhance adaptability within the farming community. Therefore, our study suggests that T_{INM} & T_{RDF} could be an option for the higher yield of rice and wheat however T_{MNF} and T_{NF3} are best options for improving soil biological health in long term. Further a long-term study on Natural farming may be taken to confirm the effect on the production



and profitability. Hence, advocating natural farming to various stakeholders is warranted, given its positive impact on soil health and cost reduction in regions favoring organic farming.

Data availability statement

The original contributions presented in the study are included in the article/supplementary material, further inquiries can be directed to the corresponding authors.

Author contributions

SD: Conceptualization, Investigation, Methodology, Resources, Validation, Writing – original draft, Writing – review & editing, Software. RS: Conceptualization, Investigation, Methodology, Resources, Validation, Writing – original draft, Writing – review & editing, Formal analysis, Supervision, Visualization. SD: Investigation, Writing – review & editing. RP: Investigation, Writing – review & editing. ND: Investigation, Writing – review & editing. PS: Investigation, Writing – review & editing. MR: Resources, Writing – review & editing. GA: Data curation, Writing – review & editing. SP: Data curation, Writing – review & editing. PR: Investigation, Writing – review & editing. MS: Investigation, Methodology, Resources, Visualization, Writing – review & editing.

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Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationship that could be constructed as a potential conflict of interest.

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