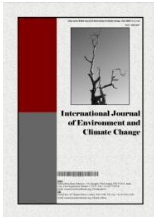

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Continuous Rice Cropping System with Integrated Use of Inorganic and Organic Sources of Nutrients for Soil Quality Improvement

P. Majhi[1], F. H. Rahman[2*] and R. Bhattacharya[2]

[1]Krishi Vigyan Kendra Jagatsinghpur, Orissa University of Agriculture and Technology, Jagatsinghpur, Odisha, India.
[2]ICAR- Agricultural Technology Application Research Institute, Bhumi Vihar Complex, Salt Lake, Kolkata- 700097, India.

Authors' contributions

This work was carried out in collaboration among all authors. Author PM designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Author FHR edited the whole draft and managed the analyses of the study. Author RB managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

Maintenance of soil quality is considered to be the key to attain sustainability in agricultural production and thus to achieve food security. In this study we tried to answer a research question **as to** whether integrated application of inorganic and organic sources of nutrients can maintain productivity and soil quality in rice-rice cropping system. To address this, total 27 physical, chemical (including organic C and soil fertility parameters) and biological (including enzyme activities) properties of a**n** acidic sandy loam soil, which were subjected to different nutrient management (NM) practices for nine years of rice-rice cropping under submergence in subtropical India were critically assessed. Seven NM practices comprised of organic [farmyard manure (FYM)], inorganic fertilizers (sources of N, P, K, S, Zn, and B) and some of their combinations were tested using randomized complete block design with four replications. Integrated use of inorganic fertilizers (NPK) and organic manure (FYM) sustained productivity of rice-rice cropping system and

**Corresponding author: E-mail: fhrahmancal@gmail.com;*

aggraded soil quality as compared to only inorganic fertilizers even with inclusion of S, Zn and B. The NPK+FYM was superior among the NM practices to improve physical, chemical and biological properties of soil. Cation exchange capacity, non-exchangeable K and microbial biomass C were screened as the most sensitive attributes for assessing soil quality. Although the present study demonstrated the positive influence of integrated NM, application of even 80-17.5-50 kg N-P-K along with 5 Mg FYM ha^{-1} in each rice season failed to maintain total K content in soil. This suggested for readjustment of dose of inorganic fertilizers and organic manure and their application schedule for adequate replenishment of K in acidic sandy loam soil under rice-rice cropping in subtropical climate.

Keywords: Rice-rice cropping; integrated nutrient management; soil organic C; soil quality.

1. INTRODUCTION

Rice (*Oryza sativa* L.) followed by rice cropping is one of the most efficient systems of rice production in south and southeastern states and medium to low land of eastern India. Growing two or three rice crops in a year in irrigated soils has been the key to national food security as well as Asia's rice supply [1]. Light-textured acidic *Inceptisols* under subtropical climate contain plenty of Fe and Al oxides and hydroxides and are inherently deficient in micronutrients i.e., Zn and B [2, 3]. Continuous supply of micronutrient-free inorganic NPK fertilizers in agriculture may further aggravate the situation of deficiencies of micronutrients [4, 5] and sometimes of secondary nutrient S [6] in soil. Sulphur deficiency in soils has been associated with poor nitrogen utilization efficiency Zhao et al. [7], while Zn deficiency causes poor seedling development and seed setting [8] and B deficiency results poor root growth [9]. Zinc and B are also necessary for pollen germination, pollen tube development and seed formation of crops [10,11]. Impairment of these biochemical activities due to inadequate supply of S and/or Zn and/or B hampers crop productivity. Therefore, application of only macronutrients (NPK) without synchronized use of those secondary or micronutrients may not be adequate to achieve potential yield of rice.

The supply of plant nutrients, particularly N, P and K through inorganic sources or organic manure into soil influences availability of not only NPK but also other essential nutrients including micronutrients and supply of any essential plant nutrient alters soil biogeochemical cycles [12,4]. Availability of essential nutrients in soil alters their concentration in plant tissues and in turn photosynthetic activity, growth and productivity of crops [13]. Variations in nutrient availability with inorganic fertilizers indirectly linked to soil C balance, but application of organic manures directly influences C balance. Changes in organic C content in soil controls soil physical, chemical (including plant nutrient availability) and biological (including activities of soil enzymes) properties [14,15,16,17,18,4]. Application of inorganic fertilizers generally reduced soil pH, whereas addition of organic amendment has been found to maintain or stabilize soil pH [19]; Naramabuye and Haynes, 2007). Inorganic fertilizers either maintain or reduce concentrations of essential plant nutrients and enzyme activities in soil. Supply of organic manure to soil either maintains or enhances their content [20, 21].

Addition of plant nutrients therefore, influences soil quality, which is one of the three components of environmental quality, other than water and air quality and is known to be a key component of sustainable agriculture [22]. The capacity of soil to support ecosystem functions for any agro-ecosystem is determined functionally by inter-linked chemical, physical and biological parameters of soil [23,24]. Soils are generally subjected to slow changes with land use and management practices and because of this, it is very difficult to record changes in soil quality unless a non-reversible damage has happened [25]. Therefore, soil quality assessment requires identification of suitable indicators, which are sensitive to changes with land use [26]. Since land management in agricultural production system usually has very little short-term effects on inherent soil properties like texture and mineralogy, other chemical and biological indicators may be more useful. Keeping these in mind, the objective of the present experiment was to evaluate the effect of integrated use of inorganic fertilizers and organic manure on soil quality after nine years of rice-rice cultivation in an *Inceptisol* located in subtropical India. To achieve this, a total of 27 physical, chemical (including organic C and plant nutrient availability) and biological (including enzyme activities) propertyes of soil under different nutrient management (NM) practices were critically examined as compared to the control (without nutrient supply) and initial soil.

2. MATERIALS AND METHODS

2.1 Experimental Site

The study was undertaken in an ongoing long-term fertilizer experiment, which was initiated in 2005 at Central Farm of Odisha University of Agriculture and Technology, Bhubaneswar (latitude 20°16' N, longitude 85°49' E, elevation of 30 m above msl) with a rice (cv., MTU-7029) -rice (cv., Lalat) cropping system under subtropical climate with an annual rainfall of 1433 mm, of which more than 85% is received during the wet season (June to September). The mean maximum and minimum temperatures were 35 and 18°C during the study period. The experimental site is a well drained medium land with an acidic, sandy loam soil classified as *Inceptisol* (*Vertic Ustochrept*).

2.2 Experiment

The experiment was laid out in a randomized complete block design with 12 treatments with four replications. Out of these 12, seven treatments viz., control (without any nutrient supply), recommended dose of NPK (NPK), 150% recommended dose NPK (NPK^), NPK+FYM, NPK+Zn, NPK+Zn+B, NPK+Zn+S were selected for th**is** study. The size of each plot was 15 m × 10 m. The recommended dose of NPK for rice in both seasons was 80-17.5-50 kg of N, P and K ha^{-1}. Farmyard manure was applied at 5 Mg ha^{-1} in each season through incorporation 15 days before final land preparation. On average, the FYM contained 165.2 g C, 8.7 g N, 3.1 g P, 6.7 g K, 3.6 g S, 28.2 g Ca, 18.6 g Mg, 1.6 g Fe, 0.9 g Mn, 88.4 mg Zn, 18.2 mg Cu and 17.3 mg B kg^{-1}. Nitrogen was applied in the form of urea (46.6% N) and partly through diammonium phosphate (21.2% N and 23.5% P) in three splits i.e., 25% as basal, 50% at 18 days after transplanting and 25% at panicle initiation stage. Entire dose of phosphorus was applied through diammonium phosphate as basal and potash was applied through muriate of potash (44.9% K) in two splits i.e., 50% as basal and 50% at panicle initiation stage. Sulphur was applied at 30 kg S ha^{-1} through gypsum ($CaSO_4$; $2H_2O$; 18.5% S) as basal. In case of Zn, ZnO (80.0% Zn) at the rate of 2 kg was added in 500 L water, stirred well and used for treating the seedlings required for one hectare by dipping their roots over night. **On the following** morning**,** seedlings were taken out and the remaining ZnO and water was spread in the respective plots. Boron was applied through foliar sprays of 0.25% borax ($Na_2B_4O_7.10H_2O$; 11.3% B) solution at 500 L ha^{-1} twice at 15 days interval during flowering.

2.3 Crop Management

Rainy (*Kharif*) season rice was transplanted in each year in the month of July and dry season rice was transplanted in the month of February after puddling with basal dose of N, P, K and S. After applying 500 mm of water, the field was puddled with power tiller. Two puddlings were then done at five days interval and finally the plots were leveled. Standing water of 3 cm was maintained throughout and 25 day old seedlings of rice were transplanted with 2-3 seedlings per hill at a spacing of 20 cm × 10 cm. Irrigation was provided to the plots intermittently as and when required in order to maintain 3 cm of standing water until 2 weeks before harvesting. Other recommended agronomic operations viz., intercultural operations and pest control measures were undertaken as and when required.

2.4 Soil Sampling and Processing and Analysis

Soil samples were collected from surface (0-15 cm) depth ten days after harvesting of dry season rice of 2015. Representative moist soil samples were collected randomly from five spots of each plot. They were bulked together to make a composite sample for each replication, then hand crushed, passed through a 2.0 mm sieve, stored at 4°C and used fresh within 24 h for estimation of soil microbial biomass C (MBC) and N (MBN), dehydrogenase, urease and acid phosphatase activity. A portion of the soil samples was air dried, milled, passed through the same sieve and used for analysis of pH, cation exchange capacity (CEC), total organic C (TOC), oxidizable organic C (OC), total N and K, non-exchangeable K and extractable plant nutrients. Another portion was used for analysis of important physical properties like sand, silt, clay and water-stable aggregates. Additional samples were taken from each plot using a core sampler (5 cm diameter, 8 cm length) for measuring bulk density (BD) and water holding capacity (WHC). Hydraulic conductivity (HC) was measured by constant head burette method. All the results were expressed on an oven-dry

(105°C) soil basis and the mean of three triplicate analyses using standard procedures.

The TOC, OC, total N content in 15 cm depth were expressed as Mg ha^{-1}; while non-exchangeable K, extractable plant nutrients as kg ha^{-1} by multiplying concentration of the respective parameters (g kg^{-1}/mg kg^{-1}) with BD and soil depth. Microbial quotient (MQ) was calculated as the ratio of MBC to TOC.

2.5 Sustainable Yield Index

Crop yield and its sustainability are the two important goal functions. For this study, crop yields and sustainability of both dry and wet seasons at ninth cropping sequence were taken as goal functions (Table **2**). Sustainability yield index (SYI) was calculated by using the formula given by Singh et al. [44].

$$SYI = \frac{Y-\sigma}{Y_{max}}$$

where, *Y* was estimated mean yield calculated over the years (2005-2015), σ was treatment standard deviation and Y_{max} was the maximum yield in that area.

2.6 Determination of Soil Quality Index

Quantitative soil quality assessment was done following the method of Andrews et al. [23] as modified by Basak et al. [26]. The procedure has two parts: i) reducing the dataset into a minimum, and ii) combining the minimum dataset (MDS) into a quality index for each treatment. The dataset (of 27 attributes) was reduced to MDS of soil quality indicators through a series of multivariate statistical analysis. Only variables with significant differences between treatment means were selected for MDS selection. Standardized principal component analysis was performed with those variables and reduced the redundancy by summing up simple correlation values among the screened variables. Representation of the screened variables towards the goal variables (crop yield, SYI) were checked and validated through computing multiple regressions. Once confirmed, the screened variables were integrated through linear weighted scoring technique into a soil quality index (SQI) for each treatment. Higher index scores were assumed to mean better soil quality.

2.7 Statistical Analysis

Rice yield and soil parameters (random variables) were subjected to analysis of variance to determine the statistical significance of treatment (fixed factors) effects. Means were separated at $p < 0.05$ using Tukey's honest significant difference. Statistical analyses were performed using SAS 9.2 [45].

3. RESULTS AND DISCUSSION

3.1 Rice Yield

Application of inorganic fertilizers, alone and in combination with FYM significantly ($p < 0.05$) increased grain yield of rice in both wet and dry seasons (Table **2**) over the control. The highest grain yield was found with NPK+FYM both in wet and dry seasons. Application of Zn in combination with NPK significantly increased rice yield, whereas NPK+Zn+B and NPK+Zn+S could not influence rice yield as compared to NPK in both the seasons.

3.2 Soil Physical Attributes

Long-term NM to rice-rice cropping system significantly ($p < 0.05$) influenced soil physical properties, except sand, silt and clay content. Integrated use of NPK+FYM had the lowest BD and the highest HC and WHC of soil among the NM. Recommended supply of N, P, K, S, Zn and B only through inorganic sources (NPK, NPK^, NPK+Zn, NPK+Zn+B and NPK+Zn+S) maintained similar HC and WHC as the initial soil, but higher values of those parameters than the control (Table **3**).

3.3 Soil Chemical Attributes

After nine years of rice-rice cultivation, soil pH decreased significantly ($p < 0.05$) with all NM practices, except NPK+FYM, as compared to its initial value (Table **4**). Supply of plant nutrients either through inorganic fertilizers and integration of inorganic fertilizers with FYM significantly increased CEC of soil over the initial status and the control and the highest value was recorded with the NPK+FYM. The highest amount of total N and available N, P, K, S, Fe, Zn and B in soil was always associated with the NPK+FYM; while the highest value of total K was in the initial soil. After nine years of rice-rice cultivation all the inorganic fertilizer treatments (NPK, NPK^, NPK+Zn, NPK+Zn+B and NPK+Zn+S) failed to maintain total N and total K as the initial soil. The inorganic fertilization, except NPK^, could only maintain comparable amount of total N and total K as the control plots. Application of inorganic fertilizers in general, maintained similar amount of available plant nutrients (N, P, K, Zn and B) as the initial soil, which were significantly higher over the control (Table **4**). However, availability of S in soil was significantly increased by its application over the control, but rice seed treatment with Zn and foliar spray of B to the crops did not make any change in available Zn and B in soil, respectively.

3.4 Soil Biological Attributes

Supply of plant nutrients (N, P, K, S, Zn and B) through inorganic sources (NPK, NPK^, NPK+Zn, NPK+Zn+B and NPK+Zn+S) significantly ($p < 0.05$) increase**d** soil MBC and MBN content over the control and **the** increase in MBC was the highest when inorganic NPK fertilizers were integrated with FYM. Similar trends were also observed for urease and acid phosphatase activity (Table **5)**, but values of dehydrogena**s**e activity with inorganic nutrient sources was lower as compared to the control. The MQ varied from 2.5 **to** 4.8%, with the mean value of 3.9%. The NM practices, except NPK, caused significant increase in MQ over the control.

3.5 Soil Organic C

Long-term rice-rice cropping without supply of nutrients (control) for nine years caused a net decrease (1.9 Mg ha^{-1} in 15 cm soil depth) in TOC content as compared to that in the initial soil (Table **4**). Rice-rice cropping with inorganic fertilization (NPK, NPK^, NPK+Zn, NPK+Zn+B and NP+Zn+S) had higher TOC (5.1 to 14.6 **Mg ha^{-1}**), but integration of NPK with FYM could increase as much as 26.2% over the initial soil. Across treatments, OC in the soil constituted 63.1 % of the TOC and followed similar trend as the TOC among the treatments compared.

3.6 Soil Quality

Among the 27 soil physical, chemical and biological attributes CEC, non-exchangeable K and MBC were selected as the MDSs of soil quality indicators through principal component analysis and validating the screened variables through multiple regression analysis taking crop yield and SYI as the goal (dependent) variables. After nine years of rice-rice cultivation with all the NM treatments except control caused an increase in SQI values in relation to the initial soil (Fig. 1). The order of the treatments in respect of magnitudes in SQI over the initial soil was NPK+FYM (0.69) > NPK^ (0.56) > NPK+Zn+S (0.43) > NPK (0.37) = NPK+Zn (0.35) > NPK+Zn+B (0.30).

Higher availability of N, P, K, S, Zn and B in soil with inorganic fertilizers (NPK, NPK^, NPK+Zn, NPK+Zn+B and NPK+Zn+S) and with combined use of NPK+FYM (Table **4**) facilitated higher removal and recovery of those nutrients by the rice crop over the control. The greater removal of nutrients helped to increase the photosynthetic activities of plants (Blevins, 1994), which improved yield of rice in both seasons (Table **3**). Supply of 30 kg S, 3.2 kg Zn and 280 g B ha^{-1} in each rice season through soil application, seed treatment and foliar spray, respectively, along with NPK (NPK+Zn, NPK+Zn+B and NPK+Zn+S) produced an average 294 kg additional rice ha^{-1} across rice season over the NPK alone. Such increase in crop yield was in agreement with those of others [46,4], who recorded significant increase in maize (*Zea mays* L.), mustard (*Brassica campestris* L.) and French bean (*Phaseolus vulgaris* L.) yield in Zn and B deficient acidic soils of northeast India. The $CaCl_2$ extractable S (9.9 and 7.6 mg kg^{-1}), DTPA extractable Zn (1.61 and 1.79 mg kg^{-1}) and hot-water extractable B (0.41 and 0.30 mg kg^{-1}) in the initial soil and the control plots, respectively, were very close to their critical values for nutrition of crops in this region [47,48,49,5]. Therefore, soils of the present experiment might be deficient in available S and/or Zn and/or B. It is established that i) S in plant metabolized into amino acids, which are important in proteins and enzymes [50], ii) Zn enhances crop growth through better seed germination, seedling growth, chlorophyll concentration [8] and iii) B improves root development [9], which result in increased nutrient uptake and crop productivity. Zinc and B are also necessary for pollen germination, pollen tube development and seed formation of crops [11,10]. In the case of NPK+FYM, supply of additional 87 kg N, 31 kg P, 67 kg K, 36 kg S, 282 kg Ca, 186 kg Mg, 16 kg Fe, 9 kg Mn, 884 g Zn, 182 g Cu and 173 g B ha^{-1} year^{-1} through FYM improved availability of those nutrients in soil (Table **4**) and in **turn** rice yield as compared to the NPK alone. Reports of integrated NM for sustainable crop harvest are available across cropping systems, soil type and climatic region [51-56,4].

Application of FYM along with NPK caused favourable changes in the soil physical properties in terms of decreasing BD and increasing HC and WHC. Production of organic acids from decomposition of applied organic amendment i.e., FYM were the possible reason for such changes in BD, HC and WHC through formation of stable soil aggregates and increasing porosity [14]. Inorganic sources of nutrient (NPK, NPK^, NPK+Zn, NPK+Zn+B and NPK+Zn+S) and integrated use of inorganic and organic sources of nutrient (NPK+FYM) increased biomass yield of rice, which could accumulate additional C through root biomass over the control [18] and could make the observed favourable changes in the physical properties (Table **3**). Effect of long-term NM on soil physical properties (i.e., BD, HC) under rice-rice cropping system as observed by Shahid et al. [57] was in agreement with our results. Reports of superiority of long-term integrated NM on soil physical properties under rice-wheat (*Triticum aestivum* L.) cropping in *Inceptisols* and *Mollisols* of subtropical north India are also there [58]; Pant and Ram [59] . Brar et al. [60] reported similar changes in soil physical properties after 36 years of wheat-maize cropping in an *Inceptisol* of north India; while Yang et al. [55] reported the same after 19 years of wheat-soybean (*Glycine max* L.) cropping in the Loess Plateau of China.

Table 1. Soil parameters analyzed

Physical
Sand, silt, clay content (international pipette method)
Bulk density by core method [27]
Hydraulic conductivity by constant head burette method [28]
Water holding capacity by Keen Raczkowski box [29]
Chemical
pH in soil water suspension of 1:2 (w/v) ratio [30]
Total organic C by combustion (TOC analyser, vario TOC select, Elementar, Germany)
Oxidizable organic carbon by Walkley and Black [31]
Cation exchange capacity by ammonium acetate method [30]
Total N by microprocessor-based Kjeldahl digestion and distillation [32]
Total K by hydroflouric acid decomposition method [33]
Non-exchangeable K by subtracting 1 M ammonium acetate extractable K from boiling 1 M HNO_3 extractable K [34]
Available N by alkaline 0.32% $KMnO_4$ extraction [35]
Available P by 0.03M NH_4F+0.025M HCl extraction [36]
Available K by neutral 1.0 M ammonium acetate extraction [37]
Available S by 0.15% $CaCl_2$ extraction [38]
Available Zn, Cu, Fe and Mn by 0.005 M DTPA+0.01 M $CaCl_2$+0.1 M TEA extraction [39]
Available B by hot-water extraction [40]
Biological
Microbial biomass C by chloroform fumigation and extraction [41]
Microbial biomass N chloroform fumigation and extraction [42]
Dehydrogenase activity by triphenylformazan [43]
Urease activity by THAM buffer method [43]
Acid phosphatase activity modified universal buffer (pH 6.5) [43]

Table 2. Effect of long-term manuring on rice grain yield and sustainability (n=3)

	Grain yield (q ha^{-1}) in 2014-15		Sustainable yield index (SYI)	
	Dry season rice	**Wet season rice**	**Dry season rice**	**Wet season rice**
Control	6.08a	18.47a	0.26	0.18
NPK	29.67b	32.28b	0.76	0.38
NPK^	32.75bc	42.41cd	0.81	0.42
NPK+FYM	36.25c	48.56d	0.89	0.51
NPK+Zn	35.71c	37.35c	0.71	0.38
NPK+Zn+B	30.00b	32.48b	0.77	0.38
NPK+Zn+S	31.75bc	36.19bc	0.73	0.39

Means followed by different letters ***in the same column*** *are significantly different at $p < 0.05$ by Tukey's honest significant difference*

Table 3. Some important soil physical properties after nine year of rice-rice cropping with different nutrient management treatments

Treatment	Sand ($g\ kg^{-1}$)	Silt ($g\ kg^{-1}$)	Clay ($g\ kg^{-1}$)	Bulk density ($Mg\ m^{-3}$)	Hydraulic conductivity ($cm\ h^{-1}$)	Water holding capacity (%)
Initial	777	93	130	1.36ab	1.52b	42.20c
Control	782	93	125	1.41b	1.37a	34.76a
NPK	773	88	139	1.35ab	1.56b	38.19ab
NPK^	771	93	136	1.35ab	1.58b	41.25bc
NPK+FYM	774	91	135	1.29a	1.63c	48.43d
NPK+Zn	783	98	119	1.37ab	1.52b	38.45b
NPK+Zn+B	782	91	127	1.34ab	1.53b	38.77b
NPK+Zn+S	775	95	130	1.37ab	1.54b	38.75b

Means (n=3) *followed by different letters* ***in the same column*** *are significantly different at $p < 0.05$ by Tukey's honest significant difference*

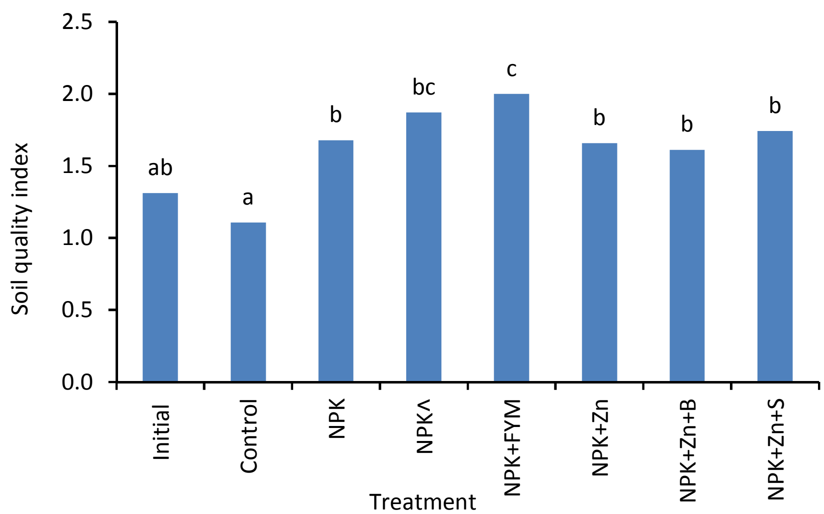

Fig. 1. Soil quality index (SQI) after nine years of rice-rice cropping under different nutrient management

Means(n=3) followed by different letters on the bars are significantly different at $p < 0.05$ by Tukey's honest significant difference

Long-term applications of acidifying fertilizers and continuous cultivation gradually develop acidic soils [61]; Kibunja et al. [62] It is known that application of urea accelerated soil acidity, whereas application of manure along inorganic fertilizers is effective in reducing soil acidity [19,63]. We observed a decrease in soil pH across the inorganic fertilizer treatments and the control from 5.80 (in the initial soil) to 5.19 after 9 years of puddled rice-rice cropping. Besides some organic acids, application of 10 Mg FYM ha^{-1} $year^{-1}$ added sufficient quantity of basic cations (i.e., 282 kg Ca and 186 kg Mg ha^{-1} $year^{-1}$), which could neutralize soil acidity [64]. This was the reason to buffer soil pH with integrated use of FYM with recommended dose of NPK as compared to the initial soil (Table **4**). Cation exchange capacity is known for its capacity to store organic C and nutrients in soil [65]. A CEC value of 14.9 c mol (+) kg^{-1} was reported to be 'optimum' for *Inceptisols* to produce 80% of potential yield of a rice-based cropping system. The mean values of CEC of the present experimental soil [3.61to 7.99 c mol (p^{+}) kg^{-1}] were far below than this 'optimum' value even with the NPK+FYM. Similar changes in CEC as observed in our study with long-term NM were also reported by others [51,60].

Table 4. Some important soil chemical properties after nine year of rice-rice cropping with different nutrient management treatments

Treatment	pH	Cation exchange capacity [c mol (p^+) kg^{-1}]	Total organic C (Mg ha^{-1})	Oxidizable organic C (Mg ha^{-1})	Total N (Mg ha^{-1})	Total K (Mg ha^{-1})	Non-exchangeable K (kg ha^{-1})	N	P	K	S	Fe	Zn	B
								Extractable essential plant nutrients in soil (kg ha^{-1})						
Initial	5.80c	5.75b	14.08ab	9.98ab	1.16b	7.20d	271a	170.3b	17.7b	39.2a	20.2b	60.2a	3.3a	0.84ab
Control	5.10a	3.61a	13.42a	8.52a	1.04ab	6.80c	301a	132.3a	7.5a	127.4b	16.2a	86.7b	3.8a	0.64a
NPK	5.24ab	6.44bc	14.89b	9.38b	0.99a	6.53ab	403b	185.8bc	9.2a	126.7b	31.4c	120.2bc	4.0a	0.67a
NPK^	5.20ab	7.13d	15.48b	9.85b	1.06ab	6.59ab	444c	198.4c	12.1ab	143.5b	36.0cd	134.7c	3.8a	0.76ab
NPK+FYM	5.93c	8.22e	17.72c	11.05c	1.17b	6.33a	450c	207.0c	23.7c	147.4b	40.5de	131.8c	4.6b	1.13b
NPK+Zn	5.19ab	6.41bc	16.20b	10.33b	0.99a	6.55ab	388b	169.2b	8.8a	118.3b	32.2cd	91.6b	3.5a	0.63a
NPK+Zn+B	5.23ab	5.82b	15.25b	9.70b	0.94a	6.37a	396b	170.7b	9.7a	122.0b	36.4cd	113.1bc	3.8a	0.87ab
NPK+Zn+S	5.21ab	6.77cd	16.00b	10.13b	0.99a	6.59b	407b	185.3bc	10.1a	120.3b	44.8e	86.7b	3.8a	0.77ab

Means (n=3) followed by different letters in the same column are significantly different at $p < 0.05$ by Tukey's honest significant difference

Table 5. Some important soil biological properties after nine year of rice-rice cropping with different nutrient management treatments

Treatment	Microbial biomass C (mg kg^{-1})	Microbial biomass N (mg kg^{-1})	Dehydrogenase activity (mg TPF kg^{-1} 24 h^{-1})	Urease activity (mg NH_4^+ kg^{-1} 2 h^{-1})	Acid phosphatase activity (mg *p*-nitrophenol kg^{-1} h^{-1})	Microbial quotient (%)
Control	158a	13.9a	179c	28.2a	229a	2.5
NPK	230b	23.4b	187c	54.4bc	279bc	3.2
NPK^	331c	31.8c	110a	59.5bc	311c	4.3
NPK+FYM	453d	44.2d	253d	81.5d	395d	4.8
NPK+Zn	336c	34.7c	157bc	56.9bc	315c	4.2
NPK+Zn+B	315c	30.7c	132ab	47.6b	318c	4.1
NPK+Zn+S	331c	34.1c	159bc	54.4bc	329c	4.2

Means (n=3) followed by different letters in the same column are significantly different at $p < 0.05$ by Tukey's honest significant difference

Lower total N and K content in NPK, NPK^, NPK+Zn, NPK+Zn+B and NPK+Zn+S treated plots as compared to the initial soil was due to increased rice yield and subsequent removal of N and K by corps, which prevented build-up of these elements in soil. Excess loss of N and K through leaching from the light-textured (sandy loam) experimental soil also caused to lower the total N and K in soil with those inorganic fertilizers. The NPK+FYM treatment that supplied 160 kg N and 100 kg K through inorganic sources and 87 kg N and 67 kg K ha^{-1} $year^{-1}$ through FYM had higher and lower total N and K content than the initial soil, respectively (Table **4**). This indicated that addition of N under the integrated treatment exceeded removal of N by the crop harvests plus its loss from soil particularly through leaching. Average decrease (14.6%) in total soil N across the inorganic fertilizer treatments from the initial soil of our study was comparable to that (20.9%) from the uncultivated/uncropped slightly acidic sandy soil (*Oxic Alfisol*) over 48 years continuous cultivation with sorghum (*Sorghum bicolor* L.) based cropping in the Sahel, West Africa [51]. Nutrient management through inorganic or its integrated use with FYM increased non-exchangeable K release (Table **4**), which could maintain greater amounts of K in exchange sites. Consequently, increased concentrations of Fe^{2+}, Mn^{2+} and NH_4^+ during flooding of acidic soil under rice-rice cropping of our experiment might displace K from the exchange complex to soil solution by establishing the equilibrium among different forms of K [66]. These were the possible reasons for increased leaching of K during prolonged flooding under double rice cropping. As a result of intense leaching of K, even NPK+FYM could not compensate for the crop removal and leaching loss of K. Therefore, it is prerequisite to replenish K in soil under such conditions to maintain positive K balance and promote efficient K management for sustainable continuous rice cultivation in acidic sandy loam soil.

The soils that received organic amendments in the form of FYM showed the highest amounts of available N, P, K, S, Fe, Zn and B. This was due to release of these elements contained in the FYM through decomposition. Besides, prolonged reduced condition under the flooded rice-rice cropping could facilitate reduction of insoluble ferric phosphate to soluble ferrous phosphate and increase the availability of P in soil [21]. Organic acids and ligands that are released during decomposition of FYM also helped to increase the availability of plant nutrients, particularly P, S and B in soil.

Soil microbial biomass C, which is very sensitive to soil management practices [67], provides an early indication of a possible degrading or aggrading effect of different management practices on soil quality [68]. The MQ values of this experiment (Table **5**) was comparable to those values (2.3 to 4.4%) reported in previous studies from this region [21,4]. The higher values of MBC and MBN under balanced NM through inorganic and organic sources of nutrients may be due to better nutritional environment as compared with the other treatments [69,17], which stimulates biological activity in soils and causes increase in MBC and MBN [70]. Comparable values of MBC and MBN with different NM for rice based cropping systems was reported earlier from the region [57]. The lowest value of MQ in the control plot indicated that the capacity of the soil for C cycling was impaired, which signifies degradation of its quality. Besides, the reduction of MQ and MBC:MBN ratio particularly with inorganic fertilization pointed out that soil microbes were under stress due to C deficiency but with abundance of N. Possibly, fungal microflora were dominant with the prevalent microbial CN ratio i.e., 9.7 to 11.4 [71] and they luxuriantly consumed N in excess of their metabolic need with inorganic fertilizers [15].

Long-term NM caused significant ($p < 0.05$) differences in soil enzyme activities; however, the changes in dehydrogenase, urease and acid phosphatase activity were dissimilar. The highest activities of these enzymes associated with the NPK+FYM were the evidences of better environment for microbial growth. Besides, observed increase in urease activity with inorganic fertilization (NPK, NPK^, NPK+Zn, NPK+Zn+B and NPK+Zn+S) indicated favourable influences of N application. Similar effect of N application in increasing urease and acid phosphatase activity like ours was reported earlier [72,15]. The result of reduced dehydrogenase activity with inorganic fertilization was in agreement with those of other researchers [21,73,74]. This indicated that inorganic N application affected dehydrogenase activity as compared to integrated nutrient sources (NPK+FYM) and N added through inorganic fertilizers were subjected to rapid transformation in soil system, removal by crop and loss through leaching instead of much influencing soil biological properties.

Organic C content in soil is generally determined by the balance between organic C inputs from plants and other soil biota over time and its losses through biogeochemical processes like decomposition, erosion and leaching, which are directly influenced by soil type, land use, and climatic zone [18]. Nine cycles of continuous rice cropping caused a net decrease in TOC content (4.2% in 15 cm depth) in the control plots as compared to the initial soil (Table **4**). This depletion in SOC was mainly due to land preparation by ploughing, which affected soil aggregates and thus facilitated oxidation of organic C [75,76]. Comparable loss of TOC (4.9%) was also reported after 36 years of continuous rice cropping under flooded condition from the same region [18]. However, greater loss of TOC even up to 60% was reported from subtropical regions [77,78] due to continuous intensive cropping without any NM. The observed lower depletion of TOC in the present experiment was because of the aquic soil moisture condition as under puddled rice-rice cropping for 7-8 months in a year and the soil being inherently low in TOC (14.08 Mg ha^{-1} in 0-15 cm depth, Table **4**). Decomposition of organic substances in soil under the prevailing anaerobic condition is generally slower than in aerated upland soil [79] and therefore, raising two rice crops in a year under puddled condition moderates the loss of TOC [80]. Supply of plant nutrients through inorganic fertilizers (NPK, NPK^, NPK+Zn, NPK+Zn+B and NPK+Zn+S) and integrated nutrient sources (NPK+FYM) caused net gain in TOC over the initial soil. This was due to increased crop C inputs in the soil through root biomass, stubble, rhizodeposition [18,81] because of an increased crop yield (Table **2**). Additional supply of C through FYM was the obvious reason for the highest increase in TOC (26.2%) under the NPK+FYM treatment over the initial soil. Carbon added through FYM is also resistant to microbial decomposition due to higher lignin and polyphenol content in FYM [77].

Cation exchange capacity, non-exchangeable K and MBC were identified as the premier contributor to influence soil quality with different NM practices. Similar to this, importance of CEC in assessing soil quality for subtropical rice-based cropping systems in *Inceptisols* was reported by Basak et al. [26]; while Benintende et al. [82] observed MBC as a good indicator for rice-based cropping system in a Vertisol. Although, total soil organic C is known as a critical soil quality indicator [83], it was found to be less sensitive to NM as compared to MBC in our case. Labile pools of soil organic C (i.e., MBC) was more sensitive to disturbance than total soil C and could better represent soil processes like nutrient cycling [84,85]. This indicated that the quality of organic C was more important than its total quantity in improving soil quality. After the ninth year of rice-rice cropping, all NM treatments tested in this experiment maintained or upgraded soil quality as compared to the initial soil and NPK+FYM was the best in the regard (Fig. 1). Similar, positive effect of integrated use of organic manure plus inorganic fertilizers across cropping system, soil order and climatic region are also there [54,86,21,4,87]. The NPK+FYM maintained availability of essential plant nutrients including micronutrients at optimum levels for nutrition of crops. Augmentation in soil physical, chemical and biological properties through long-term application of inorganic NPK along with FYM maintains higher soil quality, crop productivity and sustainability of the rice-rice agro-ecosystem in light-textured acidic *Inceptisols* under subtropical climate.

4. CONCLUSIONS

The study showed that integrated use of inorganic fertilizers and organic manure maintained the capacity of a light-textured (sandy loam) acidic soil to produce higher yield of rice-rice system under subtropical climate. The NPK+FYM was the best among the tested NM treatments to better soil quality by improving the physical, chemical and biological properties of the soil. Among the 27 soil parameters evaluated, CEC, non-exchangeable K and MBC were selected as the most sensitive to change withNM practices. Although the present study demonstrated the positive effect of integrated NM, application of even 80-17.5-50 kg N-P-K plus 5 Mg FYM ha^{-1} in each rice season failed to balance K in acidic sandy loam soil. This suggested for readjustment of rate of inorganic fertilizers and organic manure and their application schedule with regards to K for its adequate replenishment in soil under such condition without affecting K balance in rice-rice agro-ecosystem.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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