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Microbial properties of soils as affected by cropping and nutrient management practices in several long-term manurial experiments in the semi-arid tropics of India

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ABSTRACT

Microorganisms play a critical role in nutrient transformation, soil health and for sustaining the productivity of soils. Effects of long-term cropping, fertilization, manuring and their integration on microbial community were studied in soil samples from five long-term fertilizer experiments under various rainfed production systems in the semi-arid tropics (SAT) of India. Microbial population counts were analyzed by dilution plating and were in turn compared with different parameters such as soil treatments, soil type, soil microbial biomass C, soil organic C, rainfall and soil pH. The counts were high in treatments where combinations of organic and inorganic fertilizers were applied compared to control. Vertisols showed larger organic carbon levels than Alfisols. Fungal population was higher in acidic soils and in treatments under continuous inorganic fertilization treatments whereas a high number of bacteria were found in integrated use of organic and inorganic fertilizers. At most of the locations soil organic C and microbial biomass C showed significant positive ($p \leq 0.05$) correlation with microbial populations. Thus, results suggest that even under arid and semi-arid tropical conditions, regular addition of nutrients in an integrated manner could improve soil organic carbon and microbial population counts. For each production system, better carbon sequestration management practices were identified.

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1. Introduction

Microorganisms perform a key role in nutrient cycling for sustaining the productivity of the soils, because they are the source and sink for mineral nutrition and can carry out biochemical transformations (Jenkinson and Ladd, 1981). Decomposition of plant and animal residues in the soil constitute a basic biological process performed by the population of soil microorganisms. In this process, carbon is recycled as carbon dioxide (CO₂), nitrogen is converted to ammonium, and other associated elements appear in forms required by higher plants (McGill and Cole, 1981). A part of nutrients is

assimilated by microorganisms and incorporated into microbial cells (biomass). Microorganisms regulate the nutrient flow in the soil by assimilating nutrients and producing soil biomass (immobilization) and converting C, N, P and S to mineral forms (mineralization) (Jenkinson and Ladd, 1981; Wani and Lee, 1995). In the absence of soil life, all biochemical transformation cease, sustainability is endangered, and agricultural production suffers (McGill and Cole, 1981; Wani and Lee, 1995).

Low input farming systems in the tropics have adverse impact on soil organic carbon (SOC) largely through depletion of nutrients, non-return to plant residues to soil and low productivity. Legume-based systems along with appropriate

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Table 1 – Details of locations of long-term fertilizer experiments under rainfed conditions

Sl. no.	Benchmark site	State	Latitude (N)	Longitude (E)	Altitude (m)	Climate	Mean annual rainfall (mm)	Soil type
1	Anantapur	Andhra Pradesh	14.68	77.62	350	Arid	643	Alfisols
2	Bangalore	Karnataka	12.97	77.58	930	Semi-arid	924	Alfisols
3	Bellary	Karnataka	16.5	76.85	448	Semi-arid	632	Vertisols
4	Coimbatore	Tamil Nadu	11.0	77.0	426	Semi-arid	612	Vertisols
5	Solapur	Maharashtra	17.68	75.93	484	Semi-arid	742	Vertisols

soil management and nutrient management options enhance SOC contents in the tropics even without any external organic inputs (Wani et al., 2003). The soil carbon pool composed of soil organic and inorganic C play an important role in the carbon cycle. Soil organic C equilibrium is governed by a number of interacting factors such as temperature, moisture, texture, quantity and quality of organic matter, methods of organic matter application, soil tillage and cropping system. C sequestration can be augmented by increasing the quantity of organic matter returned or added to the soil, or by reducing the SOC loss by oxidation or erosion or by a combination of both. The changes in soil organic C contents are also directly associated with changes in microbial biomass carbon and biological activity in the soil. Besides living plant roots and organisms, soil microbial biomass is a living portion of soil organic matter. Maintenance of biological activity through residue management is a means for retaining organic matter and improving nutrient availability in the rainfed farming system. The response to changes in inputs of organic material is quicker in soil microbial biomass than in soil organic matter (Powlson and Jenkinson, 1981). Microbial biomass contains labile fraction of organic C and N, which are mineralized rapidly after the death of microbial cells.

Organic carbon levels increased with continuous cropping, particularly when legumes were included in the improved systems (Wani et al., 1994; Chander et al., 1997). It was observed that over a period of 5 years the change in SOC was negative under cereal–cereal sequences, whereas the SOC had increased in other cropping sequences with legume component (Singh et al., 1996). Continuous application of farmyard manure (FYM) and green manure substantially improved the organic carbon under different soils and cropping systems (Wani and Lee, 1995; Manna et al., 1997; Singh et al., 1996; Swarup, 1998). Applications of raw manures increased total organic matter, microbial biomass-N, potentially mineralizable-N and C over composted manure (Nahar et al., 2006). The tropical soils are low in organic carbon content and in principle have a large potential to sequester C through appropriate land and crop management options. Objectives of present study were to examine the relationship between soil microorganisms and management practices and to understand the link between microbial activity and C sequestration in the soils with an aim to identify the C sequestration systems in the SAT.

2. Materials and methods

Under the All India Coordinated Research Programme on Dryland Agriculture (AICRPDA), long-term fertility experiments at different centers are being conducted since 1970. The

experiments are related to predominant cropping systems and various fertility management treatments involving a combination of inorganic and organic sources (alone or together). Five long-term experimental sites selected for this study along with rainfall and soil characteristics are given in Table 1 and depicted in Fig. 1. For this study, contrasting fertility management treatments with differentiating potential for C sequestration was selected. Soil reaction, electrical conductivity (EC) particle size distribution and dominant clay mineral of initial soil samples of different locations are presented in Table 2. In all the five locations inorganic fertilizers were added in the form of NPK or as urea. Organic amendments were FYM at all sites combined and various crop residues like groundnut shells, green leaf manure at some sites. The details of the experiment and the treatments sampled for this study are described in Table 3.

2.1. Soil sample collection

Soil samples were collected from surface soil (0–15 cm) from each of the long-term experiments selected for the study during May–June 2001. In each plot, three replications with six cores (2.5 cm) in each were collected and pooled together to form a homogenous sample. The samples were stored at 4 °C and were used for the microbial analysis.

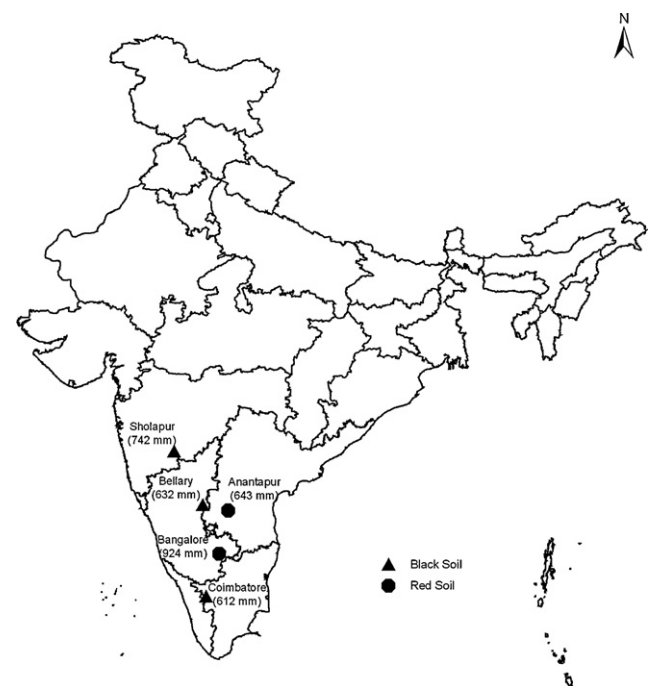
**Fig. 1 – Locations of long-term INM experiments in India.**

Table 2 – Initial soil properties of experimental sites

Location	pH (1:2)	EC (dS m ⁻¹)	Sand (%)	Silt (%)	Clay (%)	Dominant clay mineral
Anantapur	6.6	0.08	86	6	8	Kaolinite
Bangalore	5.5	0.07	74	7	19	Kaolinite
Bellary	9.0	0.29	18	22	60	Smectite
Coimbatore	8.5	2.10	56	13	31	Smectite
Solapur	8.2	0.11	6	18	76	Smectite

2.2. Biomass C

Microbial biomass was estimated by ninhydrin-reactive nitrogen extracted from soil fumigated for 5 days. Biomass C was estimated by multiplying the ninhydrin N with the factor given by Amato and Ladd (1988), which is given as under:

$$\text{biomass C} = 21 \times \text{ninhydrin reactive-N} (\mu\text{g C g}^{-1} \text{ soil})$$

2.3. Organic carbon

Organic carbon is determined by subtracting inorganic carbon from total carbon.

The total carbon (A) content of the soil samples was determined by dry combustion method using Primacs TOC analyzer. A second analysis of another set of sample (inorganic carbon (B)) is performed in the low temperature IC reactor chamber (20–150 °C). Phosphoric acid is added to the sample to acidify the inorganically bound carbon to the gaseous carbon dioxide. The flow of oxygen purges the carbon dioxide from the liquid into the IR detector to be measured again. In this way the concentration of the total organic

carbon can be calculated by subtracting the result of the low temperature measurement (B) from the high temperature (A):

$$\begin{aligned} \text{total organic carbon(TOC)} &= \text{total carbon(A)} \\ &\quad - \text{inorganic carbon(B)} \end{aligned}$$

2.4. Enumeration of microorganisms

The soil samples were enumerated for different groups of microorganisms using different growth media as indicated in Table 4.

Media prepared used according to the composition and sterilized in an autoclave. Microorganisms were enumerated using dilution and plate method on specified media plates and inoculated plates were incubated at specified temperature and duration as mentioned in Table 4. After the incubation period, the colony forming units were counted and were expressed as CFU g⁻¹ of soil. The counts were compared with different parameters such as biomass C, soil type, and different treatments in the long-term fertility management experiment.

Table 3 – Different treatments in the long-term fertility management experiment at different locations (year of sampling 2001)

Sl. no.	Location and year of experiment	Cropping system	Treatments
1	Anantapur (1985)	Groundnut	(1) Control (T1); (2) 20–40–40 kg N, P ₂ O ₅ , K ₂ O ha ⁻¹ (T2); (3) 10–20–20 kg N, P ₂ O ₅ , K ₂ O ha ⁻¹ (T3); (4) groundnut shells @ 4 t ha ⁻¹ (T5); (5) FYM @ 4 t ha ⁻¹ (T5); (6) 10–20–20 kg N, P ₂ O ₅ , K ₂ O ha ⁻¹ + groundnut shells @ 4 t ha ⁻¹ (T6); (7) 10–20–20 kg N, P ₂ O ₅ , K ₂ O ha ⁻¹ + FYM @ 4 t ha ⁻¹ (T7)
2	Bangalore (1999)	Finger millet	(1) Control (T1); (2) recommended NPK (50:50:25 kg ha ⁻¹) (T2); (3) recommended N through (GLM + FYM + CR) (T3); (4) recommended 50% N through (GLM + FYM + CR) + 50% NPK (T4)
3	Bellary (1978)	Maize–chickpea	(1) Control (T1); (2) NPK application on soil test based (T2); (3) FYM @ 5 t ha ⁻¹ year ⁻¹ (T3); (4) FYM @ 15 t ha ⁻¹ year ⁻¹ once in three years (T4); (5) NPK application on STB + FYM @ 5 t ha ⁻¹ year ⁻¹ (T5)
4	Coimbatore (1972)	Finger millet–maize–cowpea fodder	(1) Control (T1); (2) 50% optimal NPK (T2); (3) 100% optimal NPK (T3); (4) 100% optimal NPK + FYM (T4)
5	Solapur (1984)	Fallow–sorghum	(1) Control (T1); (2) 25 kg N ha ⁻¹ urea (T2); (3) 50 kg N ha ⁻¹ urea (T3); (4) 25 kg N ha ⁻¹ CR (T4); (5) 25 kg N ha ⁻¹ FYM (T5); (6) 25 kg N ha ⁻¹ CR + 25 kg N ha ⁻¹ urea (T6); (7) 25 kg N ha ⁻¹ FYM + 25 kg N ha ⁻¹ urea (T7); 25 kg N + 25 kg N ha ⁻¹ urea (T10)

GLM: green leaf manure; FYM: farmyard manure; CR: crop residue; STB: soil test based.

Table 4 – Media used, temperature, and incubation period for different microorganisms

Organism	Media	Composition (L ⁻¹)	Temperature of incubation (°C)	Period of incubation (days)
Bacteria	Nutrient agar	Agar-agar 15 g, beef extract 3 g, peptone 5 g, sodium chloride 8 g, pH 7.1 ± 0.2	25	3–5
Fungi	Potato dextrose agar	Agar-agar 15 g, potato infusion 4.0 (infusion from 200 g potatoes), dextrose 20 g, streptomycin 0.5 g, pH 5.6 ± 0.2	25	5–7
Actinomycetes	Actinomycetes isolation agar	Agar-agar 15 g, sodium caseinate 2 g, L-asparagine 0.01 g, sodium propionate 4 g, dipotassium phosphate 0.50 g, magnesium sulphate 0.10 g, ferrous sulphate 0.001 g, pH 8.1 ± 0.2	30	14

2.5. Statistical analyses

Analysis of variance (ANOVA) for soil microbial counts, biomass C and soil organic C values were analyzed using Genstat sixth edition (Payne, 2002) with randomized block design (RBD). The significant differences between the management practices, and various parameters were studied by comparing 'F' test of significance and standard error of means (Panse and Sukhatme, 1985). As a post hoc analysis Tukey's test was performed for comparison of treatment means. Simple correlation was run between the soil organic carbon and other parameters and correlation coefficient (*r*) was derived ($p \leq 0.05$).

3. Results

3.1. Alfisols

3.1.1. Anantapur

Integrated nutrient management after 16 years of cropping groundnut improved the biological parameters as compared to control. Organic carbon content of the soil increased from 3.0 g kg⁻¹ in control to 6.4 g kg⁻¹ in 50% NPK + 4 t FYM ha⁻¹ (Table 5). Though organic carbon levels increased in all the

treatments, significant ($p \leq 0.05$) increases were found only in the treatments consisting of organic manures such as FYM or groundnut shells either alone or in combination with inorganic fertilizers. Similarly soil microbial biomass carbon (MBC) improved from 115 µg C g⁻¹ in control to 150 µg C g⁻¹ with 50% NPK + 4 t groundnut shells ha⁻¹. However, the increase in microbial biomass C with various nutrient management practices was not significant. Bacteria were abundant followed by actinomycetes and fungi in this location. Bacterial population increased significantly ($p \leq 0.05$) in all the nutrient management practices when compared to control and it ranged from 4.78 (log₁₀) in control to 5.76 (log₁₀) in 50% NPK + 4 t FYM ha⁻¹ but fungi population increased significantly ($p \leq 0.05$) only in 50% NPK + 4 t FYM ha⁻¹ (4.34 log₁₀) over control (3.11 log₁₀). However, actinomycetes population increased significantly ($p \leq 0.05$) over control (3.4 log₁₀) in all the treatments.

3.1.2. Bangalore

Soil organic carbon did not increase significantly after 3 years of finger millet cropping with different nutrient management practices (Table 6). MBC levels increased significantly ($p \leq 0.05$) from 129 µg C g⁻¹ in control to 155 µg C g⁻¹ in 50% NPK + 50% N through green leaf manure + FYM + crop residue addition. Mean microbial population across the treatments

Table 5 – Microbial parameters of Alfisol soil as affected by long-term (16 years under groundnut) fertility management at Anantapur

Treatment	Soil biomass C (µg C g ⁻¹ soil)	Soil organic carbon (g C kg ⁻¹ soil)	Bacteria (log ₁₀)	Fungi (log ₁₀)	Actinomycetes (log ₁₀)	Total of microbial population (log ₁₀)
Control (T1)	115 ^a (±7.8)	3.0 ^a (±0.45)	4.78 ^a (±0.07)	3.11 ^a (±0.66)	3.40 ^a (±0.03)	4.80
20–40–40 kg N, P ₂ O ₅ , K ₂ O ha ⁻¹ (T2)	144 ^a (±7.3)	3.9 ^{ab} (±0.15)	5.11 ^b (±0.10)	3.38 ^a (±0.11)	4.40 ^c (±0.03)	5.18
10–20–20 kg N, P ₂ O ₅ , K ₂ O ha ⁻¹ (T3)	118 ^a (±2.5)	3.4 ^b (±1.43)	5.11 ^b (±0.07)	3.28 ^a (±0.07)	3.70 ^b (±0.06)	5.11
Groundnut shells @ 4 t ha ⁻¹ (T4)	138 ^a (±27.8)	4.7 ^{bc} (±1.15)	5.28 ^c (±0.02)	3.43 ^a (±0.05)	4.51 ^d (±0.07)	5.34
FYM @ 4 t ha ⁻¹ (T5)	138 ^a (±26.2)	6.3 ^d (±1.85)	5.71 ^d (±0.05)	3.46 ^a (±0.05)	4.48 ^{cd} (±0.04)	5.73
10–20–20 kg N, P ₂ O ₅ , K ₂ O ha ⁻¹ + groundnut shells @ 4 t ha ⁻¹ (T6)	150 ^a (±7.1)	4.9 ^c (±1.0)	5.43 ^d (±0.05)	3.46 ^a (±0.06)	4.54 ^d (±0.09)	5.48
10–20–20 kg N, P ₂ O ₅ , K ₂ O ha ⁻¹ + FYM @ 4 t ha ⁻¹ (T7)	145 ^a (±5.5)	6.4 ^d (±1.8)	5.76 ^e (±0.03)	4.34 ^b (±0.08)	4.45 ^{cd} (±0.06)	5.80
LSD (5%)	NS	1.4	0.12	0.43	0.09	–

Different letters in the same column indicate significant difference ($p < 0.05$) according to the Tukey test. Plus or minus values inside parentheses denotes standard error of mean.

Table 6 – Microbial parameters of Alfisol soil as affected by short-term (3 years under finger millet) fertility management at Bangalore

Treatment	Biomass C ($\mu\text{g C g}^{-1}$ soil)	Soil organic carbon (g C kg^{-1} soil)	Bacteria (\log_{10})	Fungi (\log_{10})	Actinomycetes (\log_{10})	Total of microbial population (\log_{10})
Control (T1)	129 ^a (± 9.3)	4.0 (± 1.3)	4.43 ^a (± 0.08)	2.78 ^a (± 0.07)	3.60 ^a (± 0.11)	4.49
Recommended NPK (50:50:25 kg ha^{-1}) (T2)	142 ^b (± 9.2)	4.3 (± 1.5)	5.11 ^b (± 0.10)	3.45 ^b (± 0.06)	3.96 ^b (± 0.04)	5.15
Recommended N through (GLM + FYM + CR) (T3)	146 ^{ab} (± 8.5)	4.2 (± 1.0)	5.52 ^c (± 0.04)	3.41 ^b (± 0.14)	4.20 ^c (± 0.05)	5.58
Recommended 50% N through (GLM + FYM + CR) + 50% NPK (T4)	155 ^b (± 14.5)	4.6 (± 1.2)	5.72 ^d (± 0.06)	3.40 ^b (± 0.05)	4.26 ^c (± 0.05)	5.74
LSD (5%)	24	NS	0.17	0.08	0.14	–

Different letters in the same column indicate significant difference ($p < 0.05$) according to the Tukey test. Plus or minus values inside parentheses denotes standard error of mean.

was in the order of bacteria ($5.20 \log_{10}$) > actinomycetes ($4.01 \log_{10}$) fungi ($3.26 \log_{10}$). Significant ($p \leq 0.05$) improvements were observed in all the three groups of microbes with various nutrient management practices for several years. Highest bacteria and actinomycetes population were found in INM treatments (combination of organic and inorganic) while the highest fungal population was found with recommended NPK fertilizers.

3.2. Vertisols

3.2.1. Bellary

Microbial properties of soil improved after 23 years of maize–chickpea system with different nutrient management practices (Table 7). Organic carbon content increased significantly ($p \leq 0.05$) only with soil test-based NPK + 5 t FYM ha^{-1} treatment (8.1 g kg^{-1}) as compared to control (6.5 g kg^{-1}). However, MBC increased significantly ($p \leq 0.05$) in all the treatments when evaluated against control and it ranged from $102 \mu\text{g C g}^{-1}$ (Control) to $221 \mu\text{g C g}^{-1}$ (NPK based on soil testing + 5 t FYM ha^{-1}). Bacterial population was abundant (mean $5.25 \log_{10}$) followed by actinomycetes ($4.26 \log_{10}$) and

fungi ($2.66 \log_{10}$). Significant increase ($p \leq 0.05$) in bacteria and actinomycetes population was observed with all the treatments while fungal population had not increased significantly. Total microbial population varied from $4.15 \log_{10}$ (Control) to $6.91 \log_{10}$ (NPK + 5 t FYM ha^{-1}).

3.2.2. Coimbatore

Twenty-nine years of finger millet–maize–cowpea fodder system influenced soil microbial properties considerably (Table 8). Significant increase ($p \leq 0.05$) in organic carbon content over control (5.7 g kg^{-1}) was observed in 100% optimal NPK (7.9 g kg^{-1}) and 100% NPK + FYM (8.5 g kg^{-1}). MBC also followed the similar trend where significant increase ($p \leq 0.05$) was observed with 100% NPK and 100% NPK + FYM only. However, microbial population improved significantly in all the nutrient treatments as compared to control. Bacteria were abundant (mean $4.56 \log_{10}$) followed by actinomycetes ($3.78 \log_{10}$) and fungi ($2.69 \log_{10}$).

3.2.3. Solapur

Soil organic carbon increased significantly ($p \leq 0.05$) in almost all the treatments after 17 years of fallow–sorghum system

Table 7 – Microbial parameters of Vertisol soil as affected by long-term (23 years under maize–chickpea) fertility management at Bellary

Treatment	Biomass C ($\mu\text{g C g}^{-1}$ soil)	Soil organic carbon (g C kg^{-1} soil)	Bacteria (\log_{10})	Fungi (\log_{10})	Actinomycetes (\log_{10})	Total of microbial population (\log_{10})
Control (T1)	102 ^a (± 17.5)	6.5 ^a (± 0.40)	3.73 ^a (± 0.09)	2.60 ^a (± 0.11)	3.90 ^a (± 0.11)	4.15
NPK application on soil test based (T2)	202 ^{cd} (± 24.6)	6.9 ^a (± 0.85)	5.46 ^c (± 0.03)	2.60 ^a (± 0.24)	4.18 ^b (± 0.09)	5.48
FYM @ 5 t ha^{-1} year ⁻¹ (T3)	142 ^b (± 14.9)	6.7 ^a (± 1.25)	4.77 ^b (± 0.13)	2.60 ^a (± 0.23)	4.30 ^{bc} (± 0.04)	4.90
FYM @ 15 t ha^{-1} year ⁻¹ once in 3 years (T4)	178 ^c (± 14.5)	6.8 ^a (± 1.42)	5.36 ^c (± 0.14)	2.70 ^a (± 0.09)	4.49 ^c (± 0.01)	5.41
NPK application on STB + FYM @ 5 t ha^{-1} year ⁻¹ (T5)	221 ^d (± 29.8)	8.1 ^b (± 0.46)	6.91 ^d (± 0.10)	2.78 ^a (± 0.11)	4.41 ^c (± 0.03)	6.91
LSD (5%)	36	1.0	0.17	NS	0.14	–

Different letters in the same column indicate significant difference ($p < 0.05$) according to the Tukey test. Plus or minus values inside parentheses denotes standard error of mean.

Table 8 – Microbial parameters of Vertisol soil as affected by long-term (29 years under finger millet–maize–cowpea fodder) fertility management at Coimbatore

Treatment	Biomass C ($\mu\text{g C g}^{-1}$ soil)	Soil organic carbon (g C kg^{-1} soil)	Bacteria (\log_{10})	Fungi (\log_{10})	Actinomycetes (\log_{10})	Total of microbial population (\log_{10})
Control (T1)	175 ^a (± 30.4)	5.7 ^a (± 0.10)	2.78 ^a (± 0.07)	2.30 ^a (± 0.04)	2.90 ^a (± 0.11)	3.20
50% optimal NPK (T2)	183 ^a (± 17.5)	6.7 ^a (± 0.70)	4.56 ^b (± 0.07)	2.48 ^b (± 0.07)	3.11 ^b (± 0.10)	4.57
100% optimal NPK (T3)	224 ^b (± 24.0)	7.9 ^b (± 0.15)	5.18 ^c (± 0.03)	2.78 ^c (± 0.07)	4.30 ^c (± 0.11)	5.23
100% optimal NPK + FYM (T4)	338 ^b (± 12.5)	8.5 ^b (± 0.12)	5.72 ^d (± 0.06)	3.20 ^d (± 0.11)	4.82 ^d (± 0.03)	5.76
LSD (5%)	21	1.2	0.04	0.13	0.19	–

Different letters in the same column indicate significant difference ($p < 0.05$) according to the Tukey test. Plus or minus values inside parentheses denotes standard error of mean.

consisting organic manures except of crop residue (T4) (Table 9). Overall organic carbon varied from 5.1 g kg^{-1} (control) to 11.0 g kg^{-1} (FYM + urea). However, MBC did not increase significantly in any of the treatments but integrated use of FYM + urea or crop residue + *Leucaena* maintained higher MBC than other treatments. Bacterial population showed similar trend as in the case of organic carbon whereas fungi and actinomycetes population improved significantly even in 25 kg N urea or 50 kg urea treatments. Highest total microbial population was found ($6.23 \log_{10}$) in 25 kg N-crop residue + 25 kg N ha^{-1} *Leucaena* treatment.

3.2.4. Relation between MBC and other parameters

Soil organic carbon content showed significant correlation ($p \leq 0.05$) with total microbial populations Anantapur ($r = 0.98^*$), Bangalore ($r = 0.82^*$), Bellary ($r = 0.95^*$), Coimbatore ($r = 0.98^*$) and Solapur ($r = 0.58^*$). The association between MBC and total microbial populations was also significant in all the five locations Anantapur ($r = 0.71^*$), Bangalore ($r = 0.97^*$), Bellary ($r = 0.93^*$), Coimbatore ($r = 0.80^*$) and Solapur ($r = 0.81^*$).

4. Discussion

Soils of semi-arid regions of the world are generally low in organic matter and are highly degraded. The addition of green manure improved soil biology by increasing microbial biomass and activity irrespective of management history (Stark et al., 2007). Organic matter in soil is critical for better soil health and higher soil productivity. However, maintaining or improving soil organic carbon is difficult in arid and semi-arid regions in view of rapid oxidation of organic matter due to high temperature. Regular addition of organic manures is the only way to increase soil organic matter status (Katyal et al., 2001). Repeated application of farmyard manure for 10 years developed a different microbial community compared to that amended only with chemical fertilizers (Toyota and Kuninaga, 2006). In the present study, effect of long-term addition of organic manures and inorganic fertilizers on microbial properties of some Alfisols and Vertisols under various rainfed production systems, was examined in five on going long-term fertilizer experiments. At four of the five locations were

Table 9 – Microbial parameters of Vertisol soil as affected by long-term (17 years under fallow–sorghum) fertility management at Solapur

Treatment	Soil microbial biomass C ($\mu\text{g C g}^{-1}$ soil)	Soil organic carbon (g C kg^{-1} soil)	Bacteria (\log_{10})	Fungi (\log_{10})	Actinomycetes (\log_{10})	Total of microbial population (\log_{10})
Control (T1)	126 ^a (± 24.2)	5.7 ^a (± 1.46)	5.00 ^a (± 0.04)	2.48 ^a (± 0.15)	3.83 ^a (± 0.02)	5.00
25 kg N ha^{-1} urea (T2)	133 ^a (± 23.7)	7.8 ^a (± 0.19)	5.15 ^b (± 0.11)	2.85 ^b (± 0.06)	4.41 ^d (± 0.02)	5.20
50 kg N ha^{-1} urea (T3)	136 ^a (± 39.5)	8.0 ^a (± 0.18)	5.23 ^{bc} (± 0.08)	3.00 (± 0.04)	4.41 ^d (± 0.02)	5.28
25 kg N ha^{-1} CR (T4)	127 ^a (± 36.3)	8.3 ^a (± 1.68)	5.08 ^a (± 0.07)	2.60 ^{ab} (± 0.11)	4.36 ^{cd} (± 0.06)	5.15
25 kg N ha^{-1} -FYM (T5)	140 ^a (± 42.8)	9.2 ^b (± 1.09)	5.46 (± 0.05)	3.18 ^{cd} (± 0.09)	4.30 ^c (± 0.04)	5.49
25 kg N ha^{-1} CR + 25 kg N ha^{-1} urea (T6)	143 ^a (± 36.0)	9.7 ^b (± 0.59)	5.18 ^{bc} (± 0.15)	3.1 ^c (± 0.09)	4.18 ^b (± 0.15)	5.20
25 kg N ha^{-1} FYM + 25 kg N ha^{-1} urea (T7)	164 ^a (± 2.0)	11.0 ^c (± 0.70)	5.68 ^d (± 0.02)	3.52 ^e (± 0.03)	4.40 ^{cd} (± 0.09)	5.70
25 kg N ha^{-1} CR + 25 kg N ha^{-1} <i>Leucaena</i> (T8)	162 ^a (± 10.1)	9.2 ^b (± 0.38)	6.23 ^e (± 0.08)	3.04 ^c (± 0.08)	4.11 ^b (± 0.07)	6.23
25 kg N ha^{-1} <i>Leucaena</i> (T9)	152 ^a (± 33.2)	8.6 ^b (± 1.00)	5.28 ^c (± 0.02)	2.95 ^b (± 0.05)	4.18 ^b (± 0.06)	5.30
25 kg N ha^{-1} <i>Leucaena</i> + 25 kg N ha^{-1} urea (T10)	149 ^a (± 9.5)	8.7 ^b (± 0.20)	5.34 ^c (± 0.04)	3.30 ^d (± 0.11)	4.46 ^e (± 0.05)	5.40
LSD (5%)	41	2.9	0.13	0.15	0.11	–

Different letters in the same column indicate significant difference ($p < 0.05$) according to the Tukey test. Plus or minus values inside parentheses denotes standard error of mean.

amendments were applied for 16–29 years soil organic carbon levels increased considerably due to long-term fertilization and/or manuring. At the fifth location Bangalore the soil amendments applied were only from past 3 years where the soil organic carbon was not affected by the treatments. Relationship among microbial structure, enzyme activity and N mineralization, and microbial community structure was more strongly influenced by long-term management practices than by short-term management practices (Stark et al., 2007). Increase in soil organic carbon content due to inorganic, organic, and combination underlined that the contribution of the carbon amounts directly added in manures/residues as well as to increased plant growth and residue amount returned particularly root biomass and plant root secretions to the soil following the amendments (Katyal et al., 1994). In general, Vertisols showed higher organic carbon (7.6 g C kg^{-1} soil) than Alfisols (5.5 g C kg^{-1} soil) (Fig. 2a). In the soils with the highest clay content the SOM pool responded poorly to amendments in Bellary soil (Table 7) whereas it responded more in the Solapur (Table 9). Most of the cases, regular addition of different organic manures such as FYM, groundnut shells, crop residue and green leaf manures along with chemical fertilizers improved soil organic carbon. Schulten and Leinweber (1991) reported that soil enrichment in organic matter due to application of FYM largely reflects on increase in lignin building blocks and partly reflects on increase in fatty acids. However, studies from long-term experiments on Vertisols at ICRISAT demonstrated that legume-based systems along with rainwater management and mineral fertilizer application sequestered 7.4 t C ha^{-1} than sorghum, alone which was $5 \text{ t carbon ha}^{-1}$ on application of FYM once in 2 years (Wani et al., 2003).

Different nutrient management options involving regular addition of inorganic and organic fertilizers sequestered organic carbon better than inorganic fertilizers alone. At Anantapur, $50\% \text{ NPK} + 4 \text{ t FYM ha}^{-1}$ sequestered higher carbon under continuous groundnut under arid environment. Similarly at Bangalore, combination of inorganic and organic fertilizers showed better carbon sequestration. Alfisols at Anantapur and Bangalore were acidic in reaction and application of mineral fertilizers alone neither increased SOC or microbial populations significantly. However, application of organics (FYM or residues) alone and/or in combination with mineral fertilizers increased ($p \leq 0.05$) SOC, biomass C as well as microbial populations. Different soil microbial community was established after the repeated application of chemical fertilizers and farmyard manure compared with single application of chemical fertilizers (Toyota and Kuninaga, 2006).

Except at Anantapur, improvements in MBC were more pronounced than changes in organic carbon. In general MBC maintained better association with organic carbon among soils ($r = 0.39^{\text{NS}}$ to 0.93^*) (Witter and Kanal, 1998; Powlson and Jenkinson, 1981). Soil organic carbon contents were significantly correlated with soil pH but microbial biomass C and its seasonal changes were not correlated with pH (Piao et al., 2001). Among organic sources, FYM in combination with inorganic NPK addition resulted in higher MBC except at Anantapur. Vertisols like Coimbatore and Bellary showed higher MBC than other locations (Fig. 2b). Microbial biomass

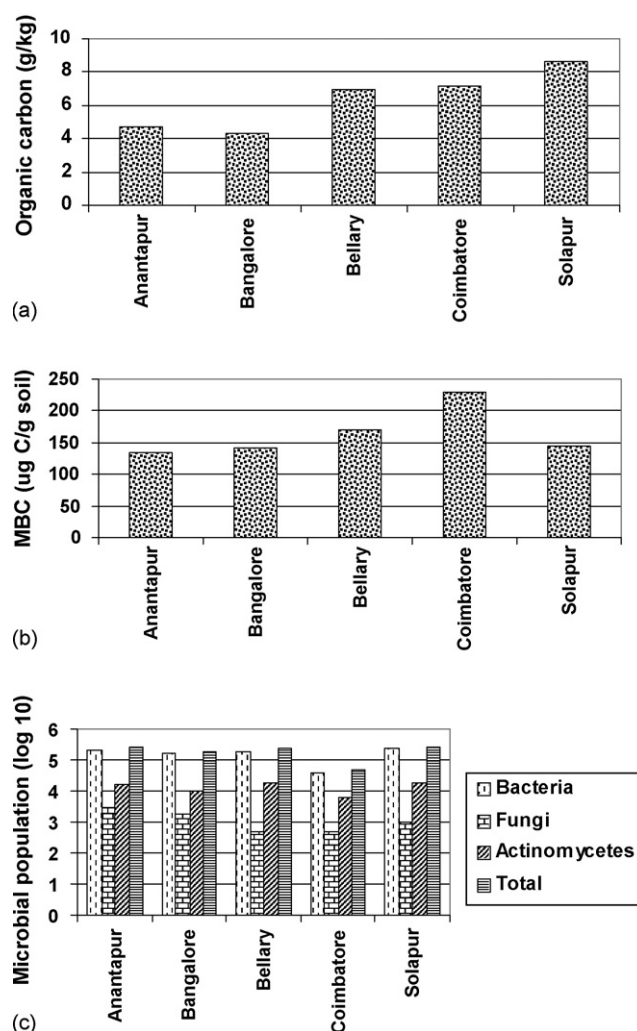


Fig. 2 – (a) Mean organic carbon, (b) microbial biomass carbon, and (c) microbial populations across the treatments in different locations.

represents a small percentage of soil organic matter, but because it is labile and dynamic in nature, it plays a significant role in nutrient cycling and ecosystem functioning (Mahmood et al., 1997; Wani et al., 2003). Type of plant cover, cropping history of soils, fertilization and manuring have been found to influence microbial biomass. The stimulatory effect of FYM on MBC is in agreement with the findings of Bolton et al. (1985). However, a smaller increase in MBC with higher FYM levels is not consistent with the results of Goyal et al. (1993), who found an increase in the stimulating effect with increasing FYM application above 15 t ha^{-1} . Burket and Dick (1998) reported that different management systems and genetic histories increased MBC content of Oregon soils.

In general, treatments consisting of different organic manures along with inorganic NPK showed better MBC than sub-optimal NPK and control. During a 100 years' long-term fertilizer experiment at Grignon, France also showed higher MBC in FYM treatment followed by mineral NPK and unfertilized control. Impact of different cropping systems significantly increased the soil MBC in two long-term fertilizer

experiments at Iowa (Moore et al., 2000). Among Vertisols, soils at Coimbatore under long-term finger millet–maize–cowpea fodder showed higher MBC followed by Bellary under maize–chickpea system as compared to fallow–sorghum system at Solapur. Inclusion of legume crop like greengram, pearl millet–wheat system increased MBC in semi-arid tropics of India (Chander et al., 1997). Similarly mean bacterial count was reported to be higher under soybean–wheat system than maize–wheat or cotton–wheat systems in southern Brazil (Balota et al., 2003).

The wide variability in the microbial population across diverse locations, soil type, production system, nutrient management practices and climatic conditions found in the present study was also observed earlier (Kumar Rao et al., 1982). Among Vertisols, Solapur with higher mean annual rainfall (742 mm) showed a slightly better microbial population followed by Bellary (632 mm) and Coimbatore (612 mm). At some sites the difference between amendments is visible on microbial numbers and not so much on soil organic carbon whereas other locations such as Solapur it was opposite. Solapur being a rain shadow region, generally rainy season crops are not grown, temperature are high and rainfall is low which would result in reduced microbial population at the time of soil sampling which was in the month of May. Similarly it was found that Vertisols having higher clay content showed larger number of microbial population (Fig. 2c). However, such associations were not found in Alfisols. Acidic soil pH recorded more counts of fungi. The biomass in the soil was dominated by fungi as microbial C:N can be seen as a measure for fungal to total microbial biomass ratio in soil (Stark et al., 2007). Similarly, in treatments with the application of inorganic fertilizers, more counts of fungi were recorded, which may be because of the change in rhizosphere soil pH. A combination of both organic and inorganic fertilizers recorded more counts of bacteria. The addition of urea, on the other hand, did not significantly affect levels of microbial biomass C and N, total C and N and microbial activity compared to the control soil (Stark et al., 2007). Among microbes, bacteria were abundant followed by actinomycetes and fungi. While reporting distribution of microorganisms in typical profile, Alexander (1971) showed similar trend of bacteria abundance. Venkateswarlu and Srinivasarao (2004) showed that both the microbial population as well as its diversity index increased in presence of FYM than fertilization and control. Similarly, number of bacterial and fungi were higher with wheat residue incorporation (Cookson et al., 1998). The size and activity of microbial populations increased with increasing inputs of crop residues (Graham and Hayens, 2005). Results suggest that soil carbon levels can be improved substantially even under arid and semi-arid regions by regular addition of organic manures along with mineral fertilizers. Higher organic carbon was found in Vertisols than Alfisols and its levels were unaffected by mean annual rainfall. Microbial population followed a similar trend and the fungal population was higher in acidic soils it was seen that organic carbon and fungi population were more in acidic soils and where only inorganic fertilizers was applied. In Alfisols, recommended NPK along with groundnut shells at Anantapur, and 50% N through GLM + FYM + CR along 50% NPK at Bangalore sequestered higher organic carbon. In Vertisols,

addition of FYM along with recommended fertilizers at Bellary and Coimbatore and 50% N through FYM 50% N through urea combinations sequestered higher organic carbon. Therefore, by choosing proper nutrient management options higher levels of organic carbon could be sequestered and thereby higher microbial population could be maintained in the soil. Higher carbon source utilization ability of the soil amended with farmyard manure may be related to different microbial community (Toyota and Kusunaga, 2006). These results bear implications on long-term productivity and sustainability of more fragile soils of arid and semi-arid regions.

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REFERENCES

- Alexander, M., 1971. *Microbial Ecology*. John Wiley and Sons, Inc, New York, NY.
- Amato, M., Ladd, J.N., 1988. Assay for microbial biomass based on ninhydrin reactive nitrogen in extracts of fumigated soil. *Soil Biol. Biochem.* 20 (1), 107–114.
- Balota, E.L., Arnold, C.-F., Andrade, D.S., Dick, R.P., 2003. Microbial biomass in soil under different tillage and crop rotation systems. *Biol. Fertil. Soils* 33, 15–20.
- Bolton Jr., H., Elliott, L.F., Papendick, R.I., 1985. Soil microbial biomass and selected soil enzyme activities: effect of fertilization and cropping practices. *Soil Biol. Biochem.* 17, 297–302.
- Burket, J.K., Dick, R.P., 1998. Microbial and soil parameters in relation to soils of diverse genesis under differing management systems. *Biol. Fertil. Soils* 27, 430–438.
- Chander, K., Goyal, S., Mundra, M.C., Kapoor, K.K., 1997. Organic matter and microbial biomass and enzyme activity of soils under different crop rotation in the tropics. *Biol. Fertil. Soils* 24, 306–310.
- Cookson, W.R., Beare, M.H., Wilson, P.E., 1998. Effects of prior crop residue management on microbial properties and crop residue decomposition. *Appl. Soil Ecol.* 7, 179–188.
- Goyal, S., Mishra, M.M., Dhankar, S.S., Kapoor, K.K., Batra, R., 1993. Microbial biomass turnover and enzyme activities following the application of farmyard manure to field soils with and without previous long-term applications. *Biol. Fertil. Soils* 15, 60–64.
- Graham, M.H., Hayens, R.J., 2005. Organic matter accumulation and fertilizer-induced acidification interact to affect soil microbial enzyme activity on a long term sugarcane management experiment. *Biol. Fertil. Soils* 41, 249–256.
- Jenkinson, D.S., Ladd, J.N., 1981. Microbial biomass in soil measurement and turnover. In: Paul, E.A., Ladd, J.N. (Eds.), *Soil Biochemistry*. Marcel Dekker, New York, USA, pp. 415–471.
- Katyal, J.C., Venkateswarlu, B., Das, S.K., 1994. Biofertilizers for nutrient supplementation in dryland agriculture: potential and problems. *Fertil. News* 39 (4), 27–32.
- Katyal, J.C., Rao, N.H., Reddy, M.N., 2001. Critical aspects of organic matter management in the tropics: the example of India. *Nutr. Cycl. Agro Ecosyst.* 61, 77–88.

- Kumar Rao, J.V.D.K., Dart, P.J., Usha Kiran, M., 1982. Cowpea group *Rhizobium* in soils of the semi-arid tropics. In: Graham, P.H., Harris, S.C. (Eds.), *BNF Technology for Tropical Agriculture*. Cali, Colombia, pp. 291–295.
- Mahmood, T., Azam, F., Hussain, F., Malik, K.A., 1997. Carbon availability and microbial biomass in soil under an irrigated wheat-maize cropping system receiving different fertilizer treatments. *Biol. Fertil. Soils* 25, 63–68.
- Manna, M.C., Hajra, J.N., Sinha, N.B., Ganguly, T.K., 1997. Enrichment of compost by bioinoculants and mineral amendments. *J. Indian Soc. Soil Sci.* 45, 831–833.
- McGill, W.B., Cole, C.V., 1981. Comparative aspects of organic C, N, S and P cycling through organic matter. *Geoderma* 26, 267–286.
- Moore, J.M., Susanne, K., Tabatabai, M.M., 2000. Soil microbial biomass carbon and nitrogen as affected by cropping systems. *Biol. Fertil. Soils* 31, 200–210.
- Nahar, M.S., Grewal, P.S., Miller, S.A., Stinner, D., Stinner, B.R., Kleinhenz, M.D., Wszelaki, A., Doohan, D., 2006. Differential effects of raw and composted manure on nematode community, and its indicative value for soil microbial physical and chemical properties. *Appl. Soil Ecol.* 34, 140–151.
- Panse, V.G., Sukhatme, P.V., 1985. *Statistical Methods for Agricultural Workers*, 4th enlarged edn. ICAR, New Delhi, India, p. 375.
- Payne, R.W. (Ed.), 2002. *The Guide to GenStat (R) Release 6.1. Part 2. Statistics*. VSN International Ltd, Oxford, UK.
- Piao, H.C., Liu, G.S., Wu, Y.Y., Xu, W.B., 2001. Relationships of soil microbial biomass carbon and organic carbon with environmental parameters in mountainous soils of southwest China. *Biol. Fertil. Soils* 33, 347–350.
- Powlson, D.S., Jenkinson, D.S., 1981. A comparison of organic matter, biomass, adenosine triphosphate and mineralisable nitrogen contents of plowed and direct-drilled soils. *J. Agric. Sci.* 97, 713–721.
- Schulten, H.C., Leinweber, P., 1991. Influence of long-term fertilization with farmyard manure on soil organic matter: characterization of particle size fractions. *Biol. Fertil. Soils* 12, 81–88.
- Singh, Y., Choudhary, D.C., Singh, S.P., Bharadwaj, A.K., Singh, D., 1996. Sustainability of rice (*Oriza sativa*)–wheat (*Triticum aestivum*) sequential cropping through introduction of legume crops and green manure crops in the systems. *Indian J. Agron.* 41, 510–514.
- Stark, C., Condron, L.M., Stewart, A., Di, H.J., Callaghan, M.O., 2007. Influence of organic and mineral amendments on microbial soil properties and processes. *Appl. Soil Ecol.* 35, 79–93.
- Swarup, A., 1998. Emerging soil fertility management issues for sustainable crop productivity in irrigated systems. In: *Proceedings of National Workshop on Long-term Soil Fertility Management through Integrated Plant Nutrient Supply System*, Indian Institute of Soil Science, Bhopal, India, April 2–4, 1998.
- Toyota, K., Kuninaga, S., 2006. Comparison of soil microbial community between soils amended with or without farmyard manure. *Appl. Soil Ecol.* 33, 39–48.
- Venkateswarlu, B., Srinivasarao, Ch., 2004. Soil microbial diversity and the impact of agricultural practices. *Indian J. Dry Land Agric. Res. Dev.* 19 (2), 97–105.
- Wani, S.P., Lee, K.K., 1995. Microorganisms as biological inputs for sustainable agriculture. In: *Thampan, P.K. (Ed.), Organic Agriculture*. Peekay Tree Crops Development Foundation, Cochin, India, pp. 39–76.
- Wani, S.P., McGill, W.B., Haugen-Koyzra, K.L., Robertson, J.A., Thurstson, J.J., 1994. Improved soil quality and barley yields with faba-beans, manure, forages, and crop rotation on a gray luvisol. *Can. J. Soil Sci.* 74, 75–84.
- Wani, S.P., Jangawad, L.S., Eswaran, H., Singh, P., 2003. Improved management of Vertisols in the semi-arid tropics for increased productivity and soil carbon sequestration. *Soil Use Manage.* 19, 217–222.
- Witter, E., Kanal, A., 1998. Characterization of the soil microbial biomass in soils from a long-term field experiments with different levels of C input. *Appl. Soil Ecol.* 10, 37–49.