

# Soil microbial biomass dynamics in grassland and agroecosystem receiving varying resource quality soil inputs in dry tropics

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## Abstract

A two year field experiments were conducted to evaluate the effect of land use conversion and management strategies variation through addition of varying resource quality soil inputs on soil microbial biomass dynamics, under rice-wheat-summer fallow crop sequence in tropical dryland agroecosystem. The treatments involved addition of equivalent amount of N through chemical fertilizer (CF) and three organic inputs, viz. *Sesbania* shoot (high quality resource, HQR), wheat straw (low quality resource, LQR) and *Sesbania*+wheat straw (mixed quality, MQR) besides control (CO) and grassland (GF). On annual mean basis, cultivation of grassland decreased the MBC (-51%) and MBN (-52%) level in soil. Relative to control, application of WS+SS considerably increased the level of MBC (+77%) and MBN (+87%) in soil, instead of SS, WS and CF. In all cultivated plots, a distinct seasonal and temporal variation in microbial biomass C and N were found contrary to GF. Both soil MBC and MBN level increased from rice to wheat period and reached maximum during summer fallow; during rice, the pattern in decreasing order was HQR>MQR>LQR>CF>CO; wheat period, MQR>LQR>HQR>CF>CO; summer fallow, MQR>LQR>HQR>CF>CO. It is concluded that cultivation exerts negative effects on the concentration of soil microbial biomass. Application of varying resource quality exogenous soil inputs maintains the microbial biomass level in soil, differentially. Addition of MOR was most effective for sustained build-up of microbial biomass in soil throughout the cycle, rather than HQR, LQR or CF. The fertility of soil in term of soil microbial biomass can be maintained by regulating the resource quality of exogenous inputs, and these results will help in designing the management strategies for sustaining long term soil fertility in these tropical dryland agroecosystem.

## Highlights

- Land use conversion from grassland to cropland significantly lowers the level of microbial biomass in soil.
- The application of single exogenous soil inputs, CF and WS resulted in much lower; SS showed marginal however, combined input SS+WS sustained higher microbial biomass throughout the annual cycle.

**Keywords:** Agroecosystem, soil microbial biomass, dryland, tropical, grassland, high quality, low quality

Soil organic matter has been considered as the reliable index of soil fertility. Microbial biomass, the active fraction (1-5%) of soil organic matter (SOM), plays a pivotal role in the availability and cycling of nutrients. The rate of nutrient fluxes in soil, primarily depends on the microbial biomass, and

thus has been used as an index of soil fertility. Due to the rapid turnover rate, soil microbial biomass has been recognized both as the transforming agent of added and native organic matter, and also as the labile reservoir for plant available N, P and S (Jenkinson and Ladd, 1981). An increase in the



size of soil microbial biomass is essential for the improvement of soil fertility. Due to its dynamic nature, the soil microbial biomass responds much more readily to changes in soil management practices as compared to total soil organic matter (Doran and Sarrantonio, 1996; Araujo and Melo, 2010).

In India ~ 63% of total arable land is under rainfed dryland farming conditions having no access to irrigation. The use of chemical fertilizer is limited in these rainfed agroecosystems due to low moisture levels. Application of organic amendments has been recommended to ameliorate soil fertility in these widely distributed drylands (Hannachi *et al.* 2015). The chemical composition of the organic inputs and the prevailing environmental conditions, govern the rate of nutrients release through microbial decomposition of organic inputs (Singh *et al.*, 2007b). Management of the microbial biomass through application of organic soil amendments has enormous potential for the management of organic matter in tropical dryland agroecosystems (Kushwaha and Singh, 2005; Singh and Singh, 1993).

Croplands are generally derived from natural grasslands in the dry tropics. Land use changes affect the dynamics of soil microbial biomass (Singh and Yadava, 2006; Dube *et al.*, 2009; Arai *et al.*, 2014), however the impact of conversion of grassland to cropland on the microbial biomass dynamics, is often conflicting. Dynamics of soil microbial biomass is widely influenced by soil amendments (Nayak *et al.* 2007; Truu *et al.*, 2008; Gong *et al.* and Lui *et al.*, 2009; Baiu *et al.*, 2012; Chang *et al.*, 2014), however, little information is available in regards to dry tropics.

In the present study, three organic inputs having contrasting chemical compositions were evaluated for their impact on the soil microbial biomass dynamics in a tropical dryland agroecosystem. These inputs included, a high quality resource (*Sesbania* shoot), low quality resource (wheat straw), and a combination of these (*Sesbania* + wheat straw) along with chemical fertilizer and the grassland.

The objective of this study was to analyze variations in the levels of soil microbial biomass C and N in the dry tropics in response to (i) cultivation of grassland to rice-based agroecosystem (ii) variations in the management strategies in terms of application

of exogenous soil amendments having contrasting chemical composition to the agroecosystem and variation within the two crop cycles and the annual cycle i.e. temporal variation

## Materials and Methods

### Study Site

The experiments were conducted in the cultivated area of the Botanical Garden of the Department of Botany, Banaras Hindu University at Varanasi (25°18' N lat. and 83°1' E long., 76 m above the mean sea level). This region has a dry tropical climate, characterized by strong seasonal variations in temperature and precipitation, including a warm rainy season (July–September), a cool winter (November–February), and a hot summer (April–June). The average annual rainfall is 1100 mm, of which approximately 80% is received during the rainy season. High temperature (24–35°C) and relative humidity (70–90%) prevail during the rainy season. In the winter season the temperature range is 4–25°C. The summer is dry and hot with a temperature range of 35 to 45°C during the day. Soil of the site belongs to the order Inceptisol, pale brown in colour, and sandy loam in texture with neutral pH.

### Experimental Design

The experimental fields have been cultivated for decades, with intermittent fallows. Since June 2002 the present experimental set up has been maintained continuously till date. The present study was undertaken for two annual cycles i.e. 2010–2011 and 2011–2012. The experimental plots were laid down in a randomized block design using three replicates per treatment. The size of experimental plots was 3m × 3m and was separated by 1 m strip. The experiment was designed to vary the quality of exogenous soil inputs carrying equivalent amount of added N (80 kg N ha<sup>-1</sup>). The following five treatments were established:

- (i) high quality organic input in form of *Sesbania aculeata* shoots having C: N::16
- (ii) low quality input in the form of wheat straw having C:N::82
- (iii) mixed quality input in the form of *S. aculeata* shoot + wheat straw having C:N::47



- (iv) chemical fertilizer (containing N:P:K::80:40:30 Kg ha<sup>-1</sup> in the form of urea, single super phosphate and muriate of potash, respectively)
- (v) control *i.e.* cropping without addition of any exogenous input along with grassland. A part of grassland was cultivated in 2002 for the establishment of our experiment.

The crop sequence was rice (*Oryza sativa* var NDR-97) as rainy season crop (July-October) followed by wheat crop (*Triticum aestivum* var Malviya 533) as winter season crop (November-March) and then the field were left fallow during summer (April-June). Both crops were directly seeded in the soil and receive natural rainfall only *i.e.* without any irrigation. The exogenous inputs were applied only once in a year before the sowing of rice crop whereas no exogenous inputs were applied for the wheat crop. *S. aculeata* was grown during summer in separate plots and were incorporated directly into the soil after chopping the upper parts of shoot into approximately 2 to 3 cm pieces.

Wheat straw was used as recycled crop residue, obtained after harvesting the wheat crop. Air dried wheat straw was cut into approximately 2 to 3 cm pieces applied to the soil. Wheat straw and *S. aculeata* shoots (corrected for moisture content) were incorporated singly or mixed as per treatment into the soil at 5 to 10 cm depth by manual hoeing, 2 days before the sowing of rice crop. Fertilizer was surface applied on the day of sowing.

The grassland was left unplanted initially (June 2002) and species common in the region (*viz.* *Dichanthium annulatum* (Forssk.) Stapf., *Cleome viscosa* L., *Corchorus capsularis* L., *Ageratum conyzoides* L., *Cyperus rotundus* L., *Clerodendrum viscosum* Vent., *Urochloa ramosa* (L.) T. Q. Nguyen and *Imperata cylindrical* L.) invaded the plot naturally. In grassland plots there was no disturbance to the soil through tillage. Grasses above 50 to 70 cm were clipped at regular intervals and the clipped biomass was left on the soil surface. No fertilization and irrigation was provided in the grassland plots.

### Soil Sampling and analysis

For the estimation of microbial biomass, soils were sampled at the vegetative, grain forming and maturity stages of each crop cycle, and once during

the summer fallow in a year covering with a total of 14 samples during the two year study. Soil samples were collected from a depth of 10 cm randomly from each replicate plot at three spots. Fresh soils were mixed to form a composite sample after removing the plant debris, and passed through 2 mm sieve. For the estimation of soil organic C and total N, soil samples were collected in April month of the first annual cycle 2010-2011.

Soil organic C was estimated by dichromate oxidation and titration method (Kalembasa and Jenkinson, 1973). In this, 0.5 g of air dried sieved soil was refluxed (20 min) by adding 20 ml of 0.5 N dichromate solution and 30 ml of acid mixture (sulphuric and phosphoric acid (5:1)). Then it was titrated against 0.5 N ferrous ammonium sulphate using phenanthroline indicator.

Total N of soil samples was estimated by the micro-Kjeldahl method (Jackson, 1973) using a Gerhardt digester and distillation unit. Soil (1g) was digested in 15 ml concentrated sulphuric acid +catalyst mixture (5g potassium sulphate + 0.5 g copper sulphate). After cooling, the volume of digested mixture was made up to 50 ml, and steam distilled with 50 ml of 40% sodium hydroxide. The distillate was collected in 40 ml of 4% boric acid indicator, and titrated against 0.1N hydrochloric acid.

For the estimation of soil microbial biomass C and N, fresh sieved soil were pre-conditioned for 7 days at room temperature in a container with 100% humidity and CO<sub>2</sub> removed (by alkali contained in vial). The container was opened for a few minute every day for aeration. Soil microbial biomass C and N was estimated by chloroform fumigation extraction method (Brookes *et al.*, 1985; Vance *et al.*, 1987), using purified CHCl<sub>3</sub> treatment, followed by 0.5 M K<sub>2</sub>SO<sub>4</sub> extraction of both fumigated and non fumigated soils. Soil microbial biomass C was estimated using the equation: MBC= 2.64 Ec (Vance *et al.*, 1987), where Ec is the difference between the amount of organic C extracted from the K<sub>2</sub>SO<sub>4</sub> extract of fumigated and non-fumigated soils, both expressed as µg C g<sup>-1</sup> dry soil. Another portion of K<sub>2</sub>SO<sub>4</sub> extracts was used for the determination of biomass N estimated as total N using Kjeldahl digestion procedure (Brookes *et al.*, 1985). The microbial biomass N was then estimated from the equation: MBN=E<sub>N</sub>/0.54 (Brookes *et al.*, 1985), where E<sub>N</sub> is the difference between the amount of

N extracted from  $K_2SO_4$  extract of fumigated and non-fumigated sets, both expressed as  $\mu\text{g N g}^{-1}$  dry soil and 0.54 is the fraction of biomass N extracted after  $CHCl_3$  fumigation. The data are expressed on oven dry soil ( $105^\circ\text{C}$ ) basis.

### MBC: MBN

Microbial C: N ratio was obtained by dividing the annual mean concentration of first and second annual cycle of soil microbial biomass C by the microbial biomass N for the grassland and all cultivated plots.

### Microbial quotient C and N

Microbial quotient C was estimated as the proportion of microbial biomass C in the soil organic C, and the fraction of microbial biomass N in the soil total nitrogen is represented as the microbial quotient N.

### Statistical analysis

Data were analyzed using SPSS (version 10.0) package on a microcomputer. All values were expressed as mean  $\pm$  one standard error. Treatment means were compared using the least square difference (LSD).

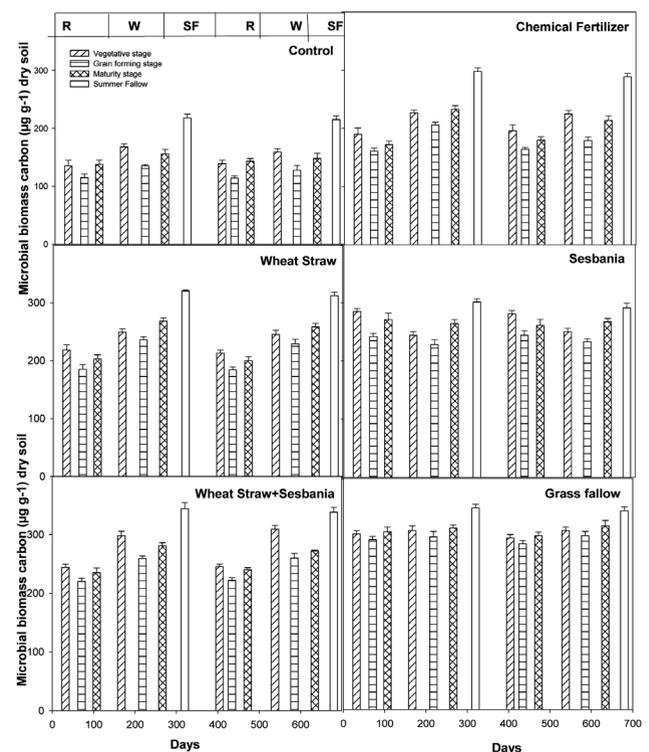
## Results and Discussion

In all cultivated plots, a marked seasonal variation in the level of soil microbial biomass C and N was observed throughout the annual cycle, which increase with low to high level from rice to wheat period and reached its maximum level during summer fallow. The variation in the level of microbial C and N were also observed within each crop cycle, among various crop growth stages i.e. the level of both microbial biomass C and N, significantly decreased from seedling to grain farming stage, and then increased till maturity during rice and wheat crop cycles. However, no such marked variation in the levels of microbial biomass C and N was found in the grassland compared to cultivated plots (Fig. 1 & 2).

### Soil microbial biomass C

The levels of soil microbial biomass C (in terms of mean of crop cycle) increased from rice to wheat crop, and reached its maximum in summer fallow in both grassland and cultivated plots except in

*Sesbania* treatments. Throughout the annual cycle, the level of soil microbial biomass C was highest in grassland compared to cultivated plots (Fig. 1). During the rice period, the level of soil microbial biomass C in grassland plots were 301 and 294  $\mu\text{g g}^{-1}$  dry soil (in first and second annual cycle respectively), which significantly decreased and reached to 129 and 132  $\mu\text{g g}^{-1}$  dry soil (in first and second annual cycle, respectively) in the cultivated control plots. However, significant increase in the level of microbial biomass C was observed after application of various soil amendments for all the treated plots compared to control plots throughout the annual cycle.



**Fig. 1:** Seasonal variation in microbial biomass carbon ( $\mu\text{g g}^{-1}$ , mean + S.E) during the various growth stages of rice wheat and summer fallow period through the two annual cycles due to application of single or combined soil amendments. Days indicates period after sowing of first rice crop Codes: R= Rice crop, W = wheat crop, SF = summer fallow

During rice period, across cultivated plots, the effect of addition of soil amendments was prominent with significant maximum level of soil microbial biomass C in *Sesbania* treatments (266 and 262  $\mu\text{g g}^{-1}$  dry soil in first and second annual cycle, respectively), followed in a decreasing order by *Sesbania* + wheat straw (233 and 235  $\mu\text{g g}^{-1}$ ), wheat straw (202 and 199  $\mu\text{g g}^{-1}$ ), and minimum in chemical fertilizer (174 and 180  $\mu\text{g g}^{-1}$ ) treatments (Table 1).

**Table 1:** Variations in soil microbial biomass C ( $\mu\text{g g}^{-1}$  soil  $\pm$  S.E) during crop and fallow periods through two annual cycles: values for rice and wheat crops are mean of three samplings during each crop cycle (2010-2012).

Crop/period	Treatments						LSD
	CO	CF	WS	SS	WS+SS	GF	
<b>2010-2011 annual cycle</b>							
Rice	129 $\pm$ 7.2	174 $\pm$ 8.4	202 $\pm$ 9.5	266 $\pm$ 12.6	233 $\pm$ 7.0	304 $\pm$ 4.4	26.5
Wheat	153 $\pm$ 9.6	222 $\pm$ 8.1	251 $\pm$ 8.2	245 $\pm$ 10.4	273 $\pm$ 12.5	307 $\pm$ 4.4	28.5
Summer Fallow	218 $\pm$ 6.6	298 $\pm$ 6.3	320 $\pm$ 2.0	301 $\pm$ 5.7	344 $\pm$ 10.3	349 $\pm$ 6.9	19.1
Annual	152 $\pm$ 7.6	212 $\pm$ 10.6	239 $\pm$ 10.1	263 $\pm$ 6.6	266 $\pm$ 9.6	311 $\pm$ 4.3	22.3
<b>2011-2012 annual cycle</b>							
Rice	132 $\pm$ 9.0	180 $\pm$ 9.2	199 $\pm$ 8.3	262 $\pm$ 10.6	235 $\pm$ 6.4	294 $\pm$ 4.4	25.6
Wheat	145 $\pm$ 9.0	206 $\pm$ 13.8	244 $\pm$ 8.3	250 $\pm$ 9.8	280 $\pm$ 13.0	310 $\pm$ 9.0	29.9
Summer Fallow	215 $\pm$ 6.6	289 $\pm$ 5.8	312 $\pm$ 6.3	291 $\pm$ 8.4	338 $\pm$ 8.5	344 $\pm$ 6.0	20.2
Annual	149 $\pm$ 7.3	203 $\pm$ 9.3	234 $\pm$ 9.4	261 $\pm$ 4.9	269 $\pm$ 9.0	308 $\pm$ 4.6	20.5

Code: CO: control, CF: fertilizer, WS: wheat straw, SS: *Sesbania* shoot, and SS+WS: *Sesbania* shoot+wheat straw and GF: grass fallow

**Table 2:** Variations in soil microbial biomass N ( $\mu\text{g g}^{-1}$  soil  $\pm$  S.E) during crop and fallow periods through two annual cycles: values for rice and wheat crops are mean of three sampling during each crop cycle (2010-2012).

Crop/period	Treatments						LSD
	CO	CF	WS	SS	WS+SS	GF	
<b>2010-2011 annual cycle</b>							
Rice	17.8 $\pm$ 1.1	26.5 $\pm$ 1.4	25.1 $\pm$ 0.9	33.5 $\pm$ 1.0	31.7 $\pm$ 1.4	36.8 $\pm$ 0.8	3.2
Wheat	18.9 $\pm$ 0.6	23.5 $\pm$ 1.6	32.7 $\pm$ 0.8	28.5 $\pm$ 1.8	36.4 $\pm$ 0.9	39.7 $\pm$ 0.6	3.0
Summer Fallow	23.5 $\pm$ 0.8	25.8 $\pm$ 1.2	36.6 $\pm$ 0.8	32.9 $\pm$ 1.0	42.3 $\pm$ 1.2	44.8 $\pm$ 1.2	2.4
Annual	18.6 $\pm$ 0.5	24.9 $\pm$ 0.6	29.3 $\pm$ 1.1	31.7 $\pm$ 0.9	33.8 $\pm$ 0.9	37.8 $\pm$ 0.7	2.3
<b>2011-2012 annual cycle</b>							
Rice	16.3 $\pm$ 1.0	25.6 $\pm$ 0.8	24.0 $\pm$ 0.5	32.7 $\pm$ 1.2	30.1 $\pm$ 1.0	35.4 $\pm$ 1.0	3.1
Wheat	17.4 $\pm$ 0.2	22.6 $\pm$ 1.0	31.2 $\pm$ 1.1	27.9 $\pm$ 1.4	34.1 $\pm$ 0.6	37.3 $\pm$ 0.6	3.0
Summer Fallow	22.8 $\pm$ 1.2	24.6 $\pm$ 0.8	35.3 $\pm$ 0.8	31.6 $\pm$ 1.2	40.8 $\pm$ 0.9	43.6 $\pm$ 0.8	3.1
Annual	17.2 $\pm$ 0.5	24.1 $\pm$ 0.4	28.1 $\pm$ 1.0	30.4 $\pm$ 0.7	33.4 $\pm$ 0.7	36.9 $\pm$ 0.7	2.0

Code: CO: control, CF: fertilizer, WS: wheat straw, SS: *Sesbania* shoot, and SS+WS: *Sesbania* shoot+ wheat straw and GF: grass fallow

In the grassland during wheat period, the levels of soil microbial biomass C was 307 and 310  $\mu\text{g g}^{-1}$  dry soil and, 349 and 344  $\mu\text{g g}^{-1}$  dry soil during summer fallow which is not significantly different from the rice period. However, in cultivated plots, the significant increase in level of microbial biomass C from rice to wheat to summer fallow period was observed. Contrary to rice period, the highest level of soil microbial biomass C was applicable to *Sesbania* + wheat straw ((273 and 280  $\mu\text{g g}^{-1}$ ) treatments rather than *Sesbania* treatments. The trend during wheat period was: *Sesbania* + wheat straw > wheat straw > *Sesbania* > chemical fertilizer > control. The same trend was also observed for the summer fallow (Table 1).

### Soil microbial biomass N

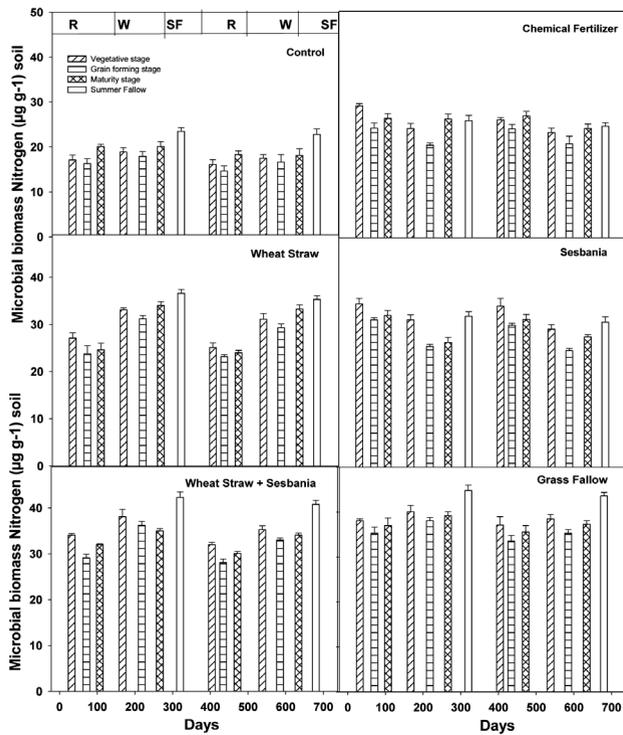
Through the annual cycle, the accumulation of soil microbial biomass N was highest in the grassland compared to cultivated plots. In grassland, the trend for seasonal variation of soil microbial biomass N was similar to that of soil microbial biomass C. The level of microbial biomass N was very similar during the cropping seasons and the crop growth stages of each crop cycle (Fig. 2). The level of soil microbial biomass N in grassland plots was 36.8 and 35.4  $\mu\text{g g}^{-1}$  dry soil (in first and second annual cycle, respectively) during rice period, and 39.7 and 37.3  $\mu\text{g g}^{-1}$  soil during wheat period, 44.8 and 43.6  $\mu\text{g g}^{-1}$  soil in summer fallow (Table 2).

**Table 3:** Organic C, total N and microbial quotient (biomass C: organic C) in soils under different treatments in the rice-wheat dryland agroecosystem; (values are mean ± S.E) of all sampling dates through two annual cycles.

Parameters	Treatments				
	CO	CF	WS	SS	WS+SS GF LSD
Organic C (%)	0.62±0.02 <sup>c</sup>	0.63±0.01 <sup>bc</sup>	0.72±0.01 <sup>b</sup>	0.65±0.06 <sup>bc</sup>	0.69±0.05 <sup>bc</sup> 0.93±0.07 <sup>a</sup> 0.057
Total N (%)	0.060±.002 <sup>c</sup>	0.070±.001 <sup>bc</sup>	0.072±.002 <sup>b</sup>	0.076±0.003 <sup>b</sup>	0.079±0.003 <sup>b</sup> 0.098±0.006 <sup>b</sup> 0.017
MBC:MBN	8.19±0.18 <sup>abc</sup>	8.53±0.27 <sup>b</sup>	8.07±0.09 <sup>ca</sup>	8.46±0.13 <sup>abc</sup>	7.79±0.07 <sup>a</sup> 8.14±0.06 <sup>abc</sup> 0.44
MBC: organic C (%)	2.43±0.02 <sup>a</sup>	3.32±0.04 <sup>c</sup>	3.29±0.03 <sup>c</sup>	4.09±0.01 <sup>b</sup>	3.88±0.02 <sup>b</sup> 3.31±0.03 <sup>c</sup> 0.21
MB N: total N (%)	3.07±0.12 <sup>d</sup>	3.51±0.05 <sup>c</sup>	4.08±0.09 <sup>b</sup>	4.10±0.08 <sup>b</sup>	4.34±0.01 <sup>a</sup> 3.87±0.08 <sup>b</sup> 0.20

Code: CO: control, CF: fertilizer, WS: wheat straw, SS: Sesbania shoot, SS+WS: Sesbania shoot + wheat straw and GF: grass fallow.

In cultivated plots, the accumulation of microbial biomass N followed the trend and pattern similar to microbial biomass C with the highest level in *Sesbania*, and the least in control treatments except that the level of microbial biomass N was higher in chemical fertilizer plots than the wheat straw treatments during rice period.



**Fig. 2:** Seasonal variations in microbial biomass nitrogen ( $\mu\text{g g}^{-1}$  soil, mean + S.E) during the various growth stages of rice wheat and summer fallow period through the two annual cycles due to application of single or combined soil amendments. Days indicates period after sowing of first rice crop Codes: R= Rice crop, W = wheat crop, SF = summer fallow

Among the cultivated plots, across treatments, the range of microbial biomass N varied from 17.8 to 33.5 and 16.3 to 32.7  $\mu\text{g g}^{-1}$  dry soil (in first and

second annual cycle, respectively) during rice period, 18.9 to 36.4 and 17.4 to 34.1  $\mu\text{g g}^{-1}$  dry soil during wheat period, and 23.5 to 42.3 and 22.8 to 40.8  $\mu\text{g g}^{-1}$  dry soil during summer fallow (Table 2). During both wheat period and summer fallow the minimum level of microbial biomass N was for control plots contrary to maximum accumulation in *Sesbania* + wheat straw treatments.

On the annual mean basis, accounting for of two study years, there was 51% decrease in the level of microbial biomass C and 52% in microbial biomass N in cultivated control plots compared to grassland (Table 1 and Table 2). Across the cultivated plots, on annual mean basis, the microbial biomass C had the trend as comparable to wheat and summer fallow period in being significantly higher in *Sesbania* +wheat straw (+77% increase over control respectively; means of two study years), and *Sesbania* (+74%) treatments relative to wheat straw, chemical fertilizer and control sets (Table1). Similar pattern was found for the microbial biomass N with a +87% rice in *Sesbania* +wheat straw treatments (increase over control; means of two study years), and +73% in *Sesbania* relative to other treatments (Table 2).

### Proportion of microbial biomass in soil organic matter

In the present study, soil organic C and total N ranged from 0.62-0.93% and 0.060-0.098% respectively, across all the treatments. Maximum content of soil organic carbon and total nitrogen (0.093% and 0.098%, respectively) was observed in grassland (Table 3). Among the cultivated plots, maximum increase in soil organic carbon (+16% increase over control) was found in wheat straw followed by wheat straw + *Sesbania* (+11.2%) owing to various soil inputs. However, total N increase



was more marked in *Sesbania* + wheat straw (+31% increase over control) followed by *Sesbania*, wheat straw and fertilizer (Table 3). The proportions of microbial biomass C and N in soil organic C and total N, respectively increased significantly for all the treatments compared to control plots. Across all the treatments, the minimum percentage of microbial biomass C in soil organic carbon C was in control (2.43%) while the maximum in *Sesbania* treatment (4.09%). However, the fraction of microbial biomass N in soil total N was highest in *Sesbania* + wheat straw treatments (4.34%) and the least in control (3.07%) (Table 3).

### **C: N ratio of soil microbial biomass**

Across all the treatments, the microbial biomass C: N ratio ranged from 7.79 to 8.53. The C: N ratio of soil microbial biomass was significantly high in chemical fertilizer (8.53) and the least in *Sesbania* +wheat straw (7.79) treatments. A significant difference was observed in the microbial biomass C: N ratio of chemical fertilizer with that of wheat straw and *Sesbania* + wheat straw treatments. Whereas the ratio was similar in *Sesbania*, grassland and control plots (Table 3).

### **Influence of resource quality on soil microbial biomass**

In the present study, on annual mean basis, the levels of soil microbial biomass carbon ranged between 152-311  $\mu\text{gg}^{-1}$  soil and 149-308  $\mu\text{gg}^{-1}$  soil in the first and second annual cycle respectively (Table 1). The present range of MBC is higher than those reported by Arunachalam and Pandey (2003) from North eastern India (175-200  $\mu\text{gg}^{-1}$  soil), Bhuyan *et al.* (2013) in agroecosystems of North east India (199-238  $\mu\text{gg}^{-1}$ ), Bowles *et al.* (2014) in arable lands of California in the U.S (67-165  $\mu\text{gg}^{-1}$ ). However, the present range of MBC is lower than range observed by Kushwaha *et al.* (2000) in rice barley agroecosystem (368-503  $\mu\text{gg}^{-1}$ ), Okur *et al.* (2009) in vineyard soils of Turkey (342-525  $\mu\text{gg}^{-1}$ ), and Gosai *et al.* (2010) for the rainfed agricultural fields in North east India (275-498  $\mu\text{gg}^{-1}$ ). A similar range of microbial biomass in soils was reported by Tu *et al.* (2006) in coastal areas of United States (134 - 371  $\mu\text{gg}^{-1}$ ), and Nayak (2007) in the flooded alluvial soils (95-318  $\mu\text{gg}^{-1}$ ).

In the present study, the accumulation of soil microbial biomass C and N found in grassland was maximum compared to cultivated plots, which could be probably attributed to higher returns of plant inputs to the soil. In grassland, no export of plant biomass from the system could have occurred in contrary to cultivated plots, and hence, the plant biomass retained by the system, and returned to the soil. In this study, there was 51% and 52% (means of two study years) decrease in the levels of soil microbial biomass C and N respectively occurred mainly on account of cultivation practices in control plots compared to grassland. The conversion of grassland to agroecosystem resulted in 16% decline in microbial biomass C in subtropical region of North India (Singh and Yadava, 2006). DuPont *et al.* (2010) reported 20% decrease in soil microbial C in non-tilled cultivated lands compared to the perennial counterpart and based such a decrease to reductions in plant inputs via root exudates and rhizodeposits.

Similar trend of reduction was also observed by Chen *et al.* (2010), He *et al.* (1997), Piao *et al.* (2000), and Accoe *et al.* (2000). Culman *et al.* (2010) reported higher levels of soil microbial biomass C and N in perennial grasslands compared to annual cultivated lands and attributed this to the input of new C at lower depths in grasslands relative to annual cropland. In contrast, Nautiyal *et al.* (2010) reported about 3 fold rise in the levels of microbial biomass C and N in the organically managed cultivated lands as compared to fallow grasslands probably due to the enhanced microbial functional diversity in the cultivated lands.

*Sesbania* shoots, a high quality resource having low lignin: N, polyphenol + lignin: N, and C: N ratio, decomposed rapidly, i.e. within 120 days (Singh *et al.* 2007). In the present study, single application of *Sesbania* resulted in +102% and 89% increase (over control, mean of two study years) in the concentration of microbial biomass C and N respectively during rice period; while a lesser increase (66% and 76%, respectively) during wheat period. Higher level of soil microbial biomass C and N, during early phase (rice crop period) of annual cycle through the application of *Sesbania*, might be due to utilization of readily available, mineralizable nutrients by microbes, and uptake by plants that could have resulted in low availability of nutrients



during the later phase (wheat crop period) of annual cycle. Considerable increase in the microbial biomass C due to incorporation of *Sesbania aculeata* in crop sequence pearl-millet-wheat in semi-arid agroecosystem is also reported by Chander *et al.* (1997). The application of *Sesbania* with Mung bean increased the microbial biomass C and N in rice wheat system of tropical soils (Tilak, 2004). Increase in the levels of soil microbial biomass C (1.94%-93.07%), and N (2.30-145.07%) respectively is also reported for in Tobacco plants, owing to application of green manure (Ye *et al.*, 2012).

On the other hand, wheat straw, a low quality resource having (C:N::81) decomposes slowly (Mohanty *et al.*, 2013) to result in the initial immobilization of nutrients into the microbial biomass, and which was remineralized and released later in the annual cycle (Singh *et al.*, 2004). This might be reason for lower soil microbial biomass C and N during early phase of annual cycle (rice crop), and higher during later phase (wheat crop) of the annual cycle. Higher microbial biomass during first crop and the marginal effect on the second crop due to single application of *Sesbania* is reported by Singh *et al.* (2004) and Singh *et al.* (2007). Aulakh *et al.* (2000) reported considerable availability of residual nutrients during the second crop. Increase in the level of microbial biomass due to straw application is also reported by several others. (Lie *et al.*, 2010; Zhao *et al.*, 2010; Azmal *et al.*, 1996; Singh and Singh, 1993; Ocio *et al.*, 1991)

Little information seems available on the effect of combined application of high and low quality inputs on soil microbial biomass dynamics in the agroecosystem. In this study, on the annual mean basis, the combined application of *Sesbania* and wheat straw maintained high level of microbial biomass C and N throughout annual cycle relative to other treatments. When *Sesbania* was added to wheat straw, it was because of the priming effect of decomposed *Sesbania* that the rate of decomposition of wheat straw increased about 2 fold compared to single application of wheat straw, thus resulting in prolonged nutrients release that in turn, supported higher microbial biomass throughout the annual cycle (Singh *et al.*, 2007).

Similar effect of high and low quality soil input (on the basis of C: N ratio) on soil microbial biomass was studied by Partey *et al.* (2014), wherein there

was higher soil microbial biomass in *Zea.mays* when applied with *Tithona.diversifolia* or *Vicia faba* (a green manure) compared to single application of *Zea mays*, and it was inferred that N supplies from the mixed input, alleviate delayed immobilization and decomposition *Zea mays*.

Impact of chemical fertilizer on the levels of soil microbial biomass was less pronounced compared to that of *Sesbania*, though both the N-rich resources were applied having equivalent N content, and this, possibly could be due to varying C contents. The tropical soils are not only limited in nutrient and moisture content but also deficient in carbon. Chemical fertilizer application may overcome the nutrient deficiency but unable to meet the C requirements and this might be reason of lower soil microbial biomass in such treatments. Contrasting responses of microbial biomass have been reported for chemical fertilizer applications. Gu *et al.* (2009), Nayak *et al.* (2007), and Graham *et al.* (2002) reported higher microbial biomass C in fertilized plots compared to unfertilized ones. Mahmood *et al.* (1997) reported that fertilization input significantly decreased microbial biomass C in wheat system but at the same time it increased in maize system. Biederbeck (1984) and Moore *et al.* (2000) reported no effect/change in microbial biomass C due to fertilizer applications. Bhattacharya *et al.* (2005) reported lower levels of microbial biomass for fertilizer treatments compared to decomposed cow manure treatments in rice crop. Higher levels of soil microbial biomass in response to organic farming rather than the conventional one (use of chemical fertilizers) were also reported by Tu *et al.* (2006), Okur *et al.* (2009) Gong *et al.* (2009) and Santos *et al.* (2011) and Chang *et al.* (2014).

### ***Influence of soil amendments on soil microbial biomass C: N ratio***

Microbial C: N ratio is the valuable index for understanding the turnover of nutrients and microbial growth efficiency in an ecosystem. According to Wardle (1992), microbial C: N reflects fungal: bacterial ratio. Greater microbial C: N ratio indicates a shift towards dominance of the fungal population in microbial biomass. Joergensen *et al.* (1995) reported that microbial C: N ratio varied from 5.2 in the arable to 20.8 in the forest soil. In most studied soils, they found an average 6.8



microbial C: N ratio. Soil amendments can change the composition of soil microflora as reflected in terms of C: N ratio of microbial biomass (Hassink *et al.* 1990). In the present study, the MBC: MBN range (7.79 to 8.53), showed the fungal dominated agroecosystem. Similar range of MBC: MBN ratio was reported by Srivastava and Lal (1994) and Hao *et al.* (2007) in tropical soils. In the intensively managed agricultural system, the mean MBC: MBN value ( $8.6 \pm 3$ ) was reported by Cleveland and Liptzin (2007) and Kellenbach (2011). However, still higher range of MBC: MBN (9.44-9.72) was reported by Singh (2013) for the dry tropical croplands.

### *Influence of soil amendments on Microbial Quotient*

The ratio of microbial biomass C to organic C and microbial biomass N to total N is known as Microbial Quotient. For predicting the dynamics and equilibrium of soil organic matter, it is the useful index (Anderson and Domsch, 1989). Generally, microbial biomass C accounts for 2-5% of total organic carbon (Jenkinson and Ladd, 1981) and microbial N represents 1-7% of total soil N (Fauci and Dick 1994; Brookes *et al.* 1985 b). Increase in the ratio of microbial biomass C to total organic C, and the microbial biomass N to total N was observed in case the organic matter was increased in system by exogenous inputs to the soil (Anderson and Domsch (1989).

Similar range of the ratio of microbial C to total C in organic soil compared to conventional soil/control soils was reported by many researchers (Srivastava and Lal, (1994), Okur *et al.* (2009), Lie *et al.* (2010). Maximum microbial quotient for C and N was found in *Sesbania*+wheat straw and *Sesbania* respectively in spite of the fact that the highest level of organic C and microbial biomass C were recorded in grass fallow treatments. Saviozzi *et al.* (2001) reported lower microbial biomass/organic C in grassland soils compared to the cultivated plots, and attributed this to greater return of crop residues (maize stalks) into the soil.

Similar findings were also reported by Powlson *et al.* (1987) and Anderson and Domsch, (1989). By using the continuous-quality model, Nilson *et al.* (2005) predicted that the microbial quotient increased due to application of straw, green manure and chemical fertilizer (calcium nitrate) relative to fallow, however,

the amount and direction of long-term changes could be misinterpreted, if predicted on the basis of short-term studies, Anderson and Domsch (1989) also concluded that on short-term experimental basis this was too early to conclude that increase in these ratios results in the accumulation of organic matter because such increases are often transient. Since it was the short period study, the shift in organic matter equilibrium as observed presently has to be viewed with caution; obviously a long-term study of combined applications is imperative.

### *Temporal variation in microbial biomass C and N*

In the present study, the effects of soil amendments on the soil microbial biomass were more pronounced than their temporal variations. However, distinct temporal variation was found in the cultivated plots. Among all the treatments, a distinct temporal variation was observed in microbial biomass C and N in both rice and wheat crop period during both the annual crop cycle despite the different climatic regimes and moisture contents. Dobosza *et al.* (1999) reported the major regulators of temporal variations were, climatic factors and crop growth and not the amount of annual organic inputs.

In the present study, for both the crop cycles a decrease in microbial C and N was observed with crop growth till the grain forming stage, and which subsequently increased at crop maturity (Fig. 1 and 2). Ghoshal and Singh (1995a) have also reported similar temporal variations in the levels of microbial biomass that declined during grain forming stage in the dryland agroecosystem. Lower microbial biomass at the grain forming stage was also reported by Singh *et al.* (2007a), and explained on the basis of a strong negative correlation between crop roots and the microbial biomass.

They suggested for a strong competition for nutrients during grain forming stage accompanied by maximum root biomass during this stage that could outcompete soil microbes thus leading to lowest soil microbial biomass. Tamilselvi *et al.* (2014) reported higher microbial biomass C and N during vegetative stage of maize which afterwards, declined till the crop harvest. Similar trends were observed by Mandal *et al.* (2007), Nayak *et al.* (2007), Li *et al.* (2012), and Kumar *et al.* (2014). Higher soil microbial biomass during the summer



fallow might be due to heavy root mortality at this stage resulting into lowering of the competition for nutrients Ghoshal and Singh (1995a). Bhattacharya *et al.* (2005) reported similar temporal trend of soil microbial biomass.

However the contrasting to increase in soil microbial biomass accompanying crop growth was also reported by (Lynch and panting (1982) and Franzluebbbers (1995). Based on the argument that increased C inputs from rhizosphere products of the soil before and during flowering, especially during fluctuating spring might increase the levels of soil microbial biomass through the vegetative stage to crop maturity.

Contrary to cultivated plots, the levels of soil microbial biomass were similar throughout the crop cycle in grassland i.e., minimal temporal variation observed could be attributed the presence of vegetation continuously throughout the crop cycle. Among these *Dichanthium annulatum* (Forssk.) Stapf., *Cleome viscosa* L., *Corchorus capsularis* L., *Ageratum conyzoides* L., *Cyperus rotundus* L., *Clerodendrum viscosum* Vent., *Urochloa ramosa* (L.) T. Q. Nguyen, and *Imperata cylindrical* L.) were the dominant components of vegetation in the grassland. Such grasses and forbes having elaborate root systems were present almost throughout the year except summer.

As a result, the competition between plant roots and microbes for nutrients were not so pronounced at any phase of the annual cycle, contrary to conditions in cultivated plots. During the late wheat period and summer fallow the vegetation started drying due to lack of availability of soil moisture and rise in temperature, and this possibly lowered the competition between microbes and roots and supported microbial biomass. Gayston *et al.* (2001) also reported little variation in the grassland. However, Patel *et al.* (2010) reported distinct variations in the levels of microbial biomass between rainy and winter season. Moisture availability and temperature were reported to be the major drivers of seasonal variation in microbial biomass in grasslands (Bell *et al.* 2008).

## Conclusion

Land use conversion from grassland to cropland decrease the level of microbial biomass in soil. Application of different resource quality exogenous

inputs (having equivalent amount of N but variable in carbon content) influences the content of microbial biomass differentially in soil. High quality resource inputs promoted highest level of microbial biomass during early phase of annual cycle however; low quality resource inputs delayed microbial biomass accumulation to later phase. The mixed resource quality inputs sustained higher microbial biomass throughout the annual cycle, benefiting both crops. It is necessary to standardize management practices using diverse organic inputs in appropriate combination with a view to sustaining enhanced soil fertility for a longer period, which in turn shall help in sustainable agroecosystem.

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