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Farm Management to Control of Soil Microbial Density and Metabolic Activities in Rice-Rice Agroecosystem

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Abstract: Soil microbial communities are likely to be influenced by agricultural practices, which change the soil environment. One of such practice is the use of organic amendment instead of conventional chemical inputs. This study reported the status of important available soil nutrients i.e., OC, N, P and K along with bacterial and fungal population at different soil depths in organically as well as conventionally managed tropical agroecosystems. Significantly higher ($P<0.01$) bacterial and fungal population was observed in organic in comparison to conventional farming systems. There was significant difference in the microbial density of two agroecosystems, the organic one having more density than of conventional one. The differences in microbial population were also found significant between soil depths and between seasons. In both cultivation conditions, there was gradual decrease of microbial density with increase in the soil depth in most part of the year excepting summer months, whereas inner soil layers were healthier in microbes than the surface soil. The variations in microbial population have been attributed to the availability of soil macro nutrients. The organic and inorganic nutrient levels were higher in the surface soil than in the inner layers in both the agroecosystems. The soil enzymes, viz., activities of amylase, invertase and cellulase, could be well correlated with the microbial density.

Key words: Organic farming · Conventional farming · Bacteria · Fungi · Enzymatic activities

INTRODUCTION

The productivity of the rice agroecosystem has been found to be significantly influenced by the density, diversity and metabolic efficiency of soil microflora. Soil enzymes catalyze the biochemical reactions responsible for degradation of soil organic matters (SOC) and nutrient cycling in the soil subsystems [1, 2]. The chemical changes brought about by these exoenzymes in the soil are important for fertility and plant growth. The soil enzyme complex represents a completely separate system consisting of enzymes associated with dead soil components [3]. Soil enzymes are important for life processes of microorganisms and stabilization of soil structure [4]. Microbial biomass and soil enzyme activities can potentially provide an integrated biological assessment of soil quality [1, 5]. The relationships between soil organic matter, microbial biomass and microbial activity have been proposed as indicators of soil maturity [6]. Organic farming is an ecofriendly system of farming which can maintain soil health in terms of soil biological fertility and productivity and maintain a balance among the organic, inorganic and biological components of soil. The favourable effect of various components of organic farming over conventional farming systems or integrated nutrient management practice is to be considered holistically rather than looking for short term benefits [7]. Limited information is available on the magnitude of seasonal as well as depth wise changes in soil enzyme activity and microbial population of conventional and organic agroecosystems especially in the widely prevalent rice based cropping system of tropical region. The exact relationship between the microbial activities and farm practices are also needed to be developed. This paper presented the impact of the soil microbial density on the soil enzyme activities as well as on the soil nutrient status with variation of farming practices.

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MATERIALS AND METHODS

Sampling: The study was conducted during March 2007 to January 2009 in three different seasons i.e. summer (March-May), rainy (July-September) and winter (November-January). Soil samples were collected from rice fields under both organic and conventional farming systems located in Khurda district of Orissa state and maintained by the Orissa University of Agriculture and Technology, Bhubaneswar. Sampling was done at an interval of 15 days during the entire period of study. The sites were characterized by a climate with most rainfall occurring during rainy months (July-September). The ambient temperature average ranged from a minimum of 11.1°C to a maximum of 44.6°C and annual normal rainfall of 1449.1 mm. Soil samples were collected from top 20cm of soil and were segregated into five depth classes at 4 cm intervals. Collection was made in sterilized polythene bags using a steel corer and each sample was divided into three replicates. The replicates were labeled and left for isolation and enumeration of bacteria and fungi within 24 h of collection. The enzymatic activity of soil samples were measured immediately after collection.

Major Soil Macro Nutrients Studies: Organic carbon (OC) was estimated titrometrically [8], mineralisable nitrogen (N) was done by the method of Subbaiah and Asija [9]. Available Phosphorus (P) and Potassium (K) were carried out by Olsen [10] and Ammonium acetate flamephotometry method [11], respectively.

Isolation of Bacterial and Fungal Population: The isolation of bacteria and fungi was done by spread plate method using Nutrient Agar and Potato Dextrose Agar, respectively. Soil microbial population was estimated by dilution plate method [12]. The inoculated plates were incubated at 37°C for 24 h for bacteria and at 25°C for 72 h for fungi. The colony was counted with the help of a digital colony counter.

Enzymatic Activity: The activity of carbohydrate enzymes viz., amylase, cellulase and invertase were measured spectrophotometrically with the help of a uv-vis spectrophotometer (Perkin-Elmer EZ-201, Überlingen, Germany) using glucose as standard [13]. The substrates used were starch, carboxymethyl cellulase and sucrose (each at 1% concentration) for the activity of amylase, cellulase and invertase, respectively.

Statistical Analysis: The samples were collected in triplicates from 5 plots each of fifteen zones. The replicates of one sample zone were averaged as one replicate. The statistical analysis of all the data has been done using M Stat C Software (Michigan State University, USA). The data were analyzed through ANOVA (at $P = 0.05$ levels) among the seasons and the depths. Correlation among bacterial and fungal populations as well as the activity of the enzymes was made following standard statistical procedures [14].

RESULTS AND DISCUSSION

The bacterial (x 10^4 cfu /g dry soil) and fungal (x 10^3 cfu /g dry soil) population were consistently higher in organically managed soil in comparison to conventional rice field soil in all the seasons and all soil depths (Figs. 1 and 2). There was significant depth wise variation of the microbial density, which varied in trend with the seasons. The highest bacterial and fungal count was observed in top 4 cm soil layer in winter and rainy seasons in both the farming systems while in summer the innermost soil layer of the fields was denser in microbes. In all seasons and soil layers the microbial density of organic fields was significantly higher than of conventional fields. The difference in annual average of microbial density of two farming systems was more prominent in the top 4 cm of soil than in other soil depths. The bacterial and fungal density of conventionally managed rice field soil were 65 and 84% of that of organically managed field, respectively, indicating that the bacteria are more sensitive to the farm practices than of soil fungi. Remarkable difference of the microbial population was observed between the surface and the inner layers of soil. The bacterial population reduced at a faster rate with increase of soil depths than of the fungal population. While in organic fields the depth wise variation of bacterial density was less pronounced that of conventional fields, the reverse trend was reported with respect to soil fungi.

The farming practice could cause significant change of the soil nutrient status and the soil organic carbon (SOC) content during the observation period (Tables 1 and 2). Though in both organic and conventional farming systems the SOC level decreased with increase of soil depth, the rate of reduction in the organic field was significantly higher than of conventional one. Corresponding changes were also reported with respect to soil nutrient levels.

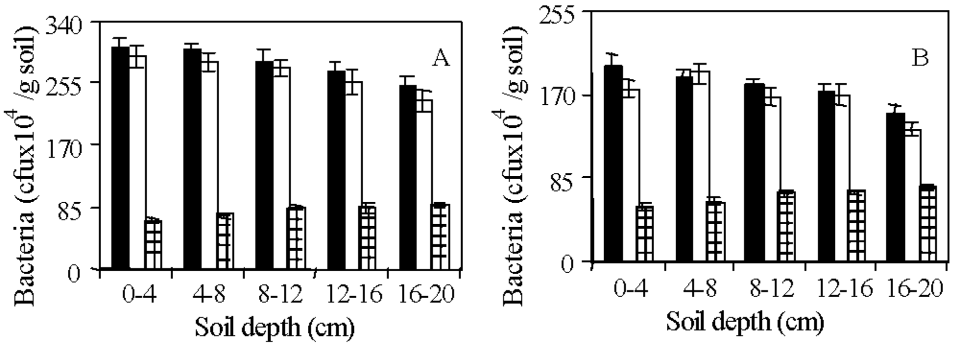

Fig. 1: Depth wise variation in bacterial population (x 10^4 cfu/g soil) in (A) organic and (B) conventional farming system. Histograms: *full*- winter; *empty*- rainy; *bricks*-summer.

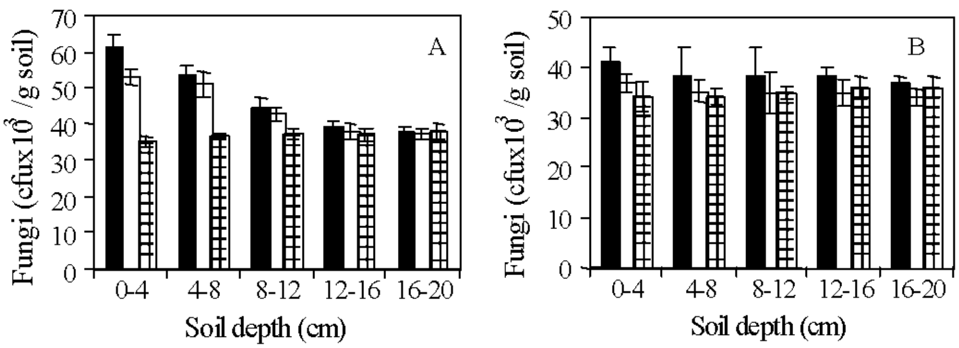

Fig. 2: Depth wise variation in fungal population (x10^3 cfu/g soil) in (A) organic and (B) conventional farming system. Histograms: *full*- winter; *empty*- rainy; *bricks*-summer.

Table1: Depth wise variation of soil organic carbon organic carbon (SOC: g/kg), mineralizable nitrogen (MN: kg/ha), available phosphorus (AP: kg/ha) and available potassium (AK: kg/ha) in organic (O) and conventional (C) farming systems

	SOC		MN		AP		AK	
Depth range (cm)	O	C	O	C	O	C	O	C
0-4	9.12±1.04	4.70±0.48	156.61±7.34	131.825.09	30.11±4.78	21.12±2.42	168.2±7.66	137.64±11.21
4-8	8.39±0.81	4.66±0.35	150.11±7.12	130.71±4.51	27.56±2.91	18.14±2.27	144.4±9.46	124.21±13.38
8-12	8.28±0.74	4.52±0.41	146.32±4.04	132.35±6.11	25.56±2.01	18.02±2.03	133.1±11.51	114.47±5.50
12-16	7.84±0.51	4.38±0.31	138.23±6.01	128.13±4.98	24.11±2.40	16.23±2.52	119.3±12.89	101.22±9.01
16-20	7.69±0.48	4.21±0.43	136.41±5.70	126.26±4.97	21.89±1.50	16.17±1.02	109.7±6.72	98.52±8.35

Table 2: Seasonal variation of soil organic carbon organic (SOC: g/kg), mineralizable nitrogen (AN: kg/ha), available phosphorus (AP: kg/ha) and available potassium (AK: kg/ha) in organic (O) and conventional (C) farming systems for three crop fields

	SOC		MN		AP		AK	
Season	O	C	O	C	O	C	O	C
Winter	9.02±0.73	4.83±0.24	148.84±10.61	135.44±6.32	29.73±3.19	19.81±2.84	157.24±14.76	131.15±15.78
Summer	7.66±0.29	3.98±0.18	139.26±6.98	125.52±4.34	20.93±2.49	15.64±1.52	116.19±9.59	100.44±6.56
Rainy	8.12±0.69	4.66±0.19	146.71±5.51	128.57±2.35	26.87±3.83	18.32±2.61	131.43±12.45	113.92±11.71

Araujo *et al.* [15] observed that SOC levels not only facilitate the microbial activity but also lower bulk density. The SOC levels in organic farming systems steadily increases with continuation of the farm practice causing positive soil conditioning. Singh *et al.* [16] observed that organic manuring along with farmyard manure and green manure without application of any inorganic fertilizers proved quite effective in producing high rice yield. However, the differences of the mineralizable nitrogen (MN), available potassium (AK) and available phosphorus (AP) between the two farming systems were found less than of the SOC indicating that the

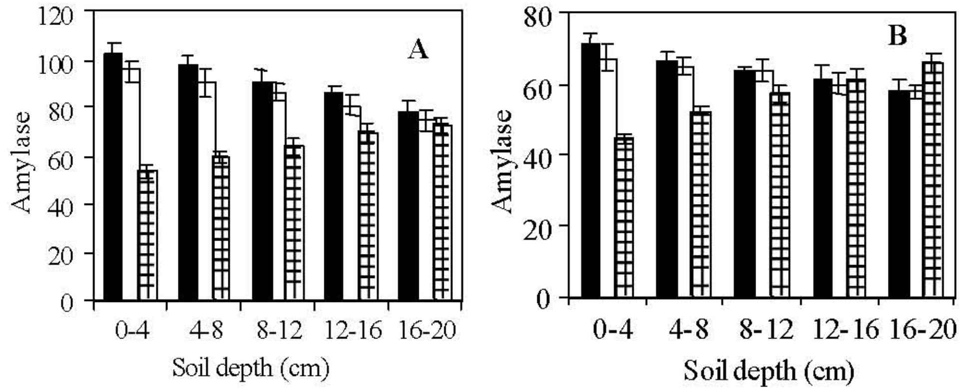

Fig. 3: The activity amylase (µg glucose released/g soil) in (A) organic and (B) conventional farming system with change in soil depth. Histograms: *full*- winter; *empty*- rainy; *bricks*-summer.

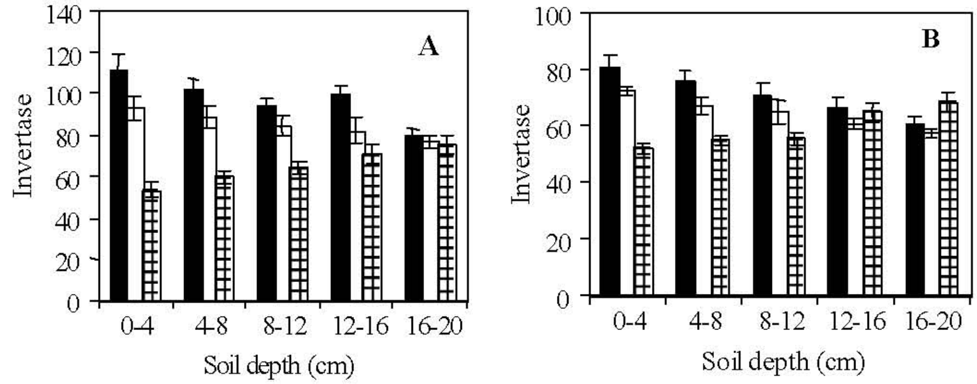

Fig. 4: The variation of invertase activity (µg glucose released/g soil) in (A) organic and (B) conventional farming system with change in soil depth. Histograms: *full*- winter; *empty*- rainy; *bricks*-summer.

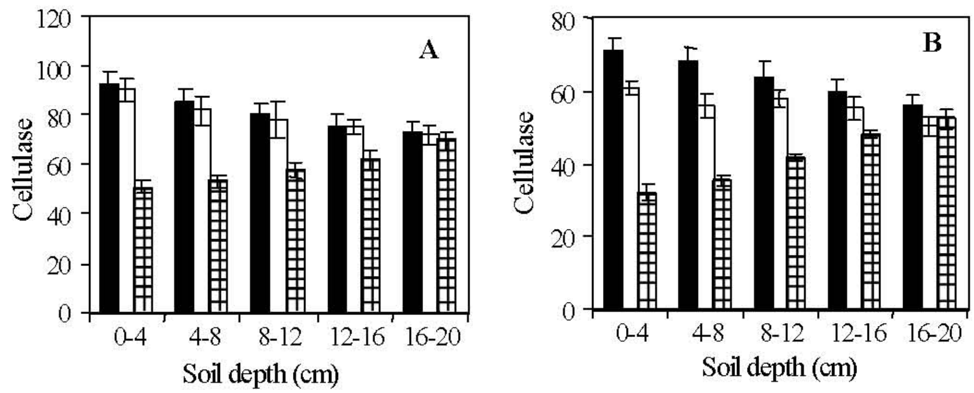

Fig. 5: The activity cellulase (µg glucose released/g soil) in (A) organic and (B) conventional farming system with change in soil depth. Histograms: *full*- winter; *empty*- rainy; *bricks*-summer.

conventionally managed soil could enjoy an acceptable level of nutrients despite of significantly low quantity of SOC due to the amendment of chemical fertilizers.

Irrespective of seasons, the activities (µg glucose /g soil) of amylase (Fig 3), invertase (Fig 4) and cellulase (Fig 5) progressively declined with increasing soil depth and the innermost soil layer exhibited minimum activity in winter and rainy seasons, but a reverse trend was observed in summer. There was no significant difference between the organically and conventionally managed soil with respect to the rate of reduction of the activity of invertase and cellulase with increase in the soil depth. However, the activity of amylase was found reduced at a higher rate in conventional soil than in the organic one.

The enzyme activities were the highest in winter and the lowest in summer. Further, organic fields exhibited higher enzyme activity during all the season in comparison to conventional one.

Generally the microbial load is generally higher in the surface soil as compared to inner layer as surface are organically richer than the inner layers, which was observed in both the farming systems in the present case. Further, overall reduction in microbial population in the lower soil depths has been attributed to lesser amounts of minerals, oxygen availability and higher carbon dioxide concentration [17]. Our results are in agreement with the conclusions of Jha *et al.* [18] and Tangjang *et al.* [19] who recorded higher bacterial population during post rainy (winter) seasons. The minimum bacterial population in both the farming systems was observed in summer which is otherwise critical for the growth and activity of microbes. Increase in number of microbial population in summer towards deeper soil layers as seen from our results corroborates that of Classen *et al.* [20] who pointed out that during hot summer months, the sub layer of soil occasionally harbours more fungal populations caused by temperature and moisture regimes than the topsoil layer. Higher fungal population during winter supported the findings of other workers [21, 22] which perhaps is due to prevailing favourable moisture and temperature settings during the post monsoon (winter period) and the accumulation of humus during monsoon which may have enhanced the colonization of soil microbes in post monsoon period. Irrespective of season organic soils has higher bacterial and fungal population over conventional soil.

Earlier studies have shown that the activity of soil enzymes can be used as a sensitive indicator of changes in soil biological activity in response to various soil management practices. Amylase activity is affected by organic matter content and cultural practices [23]. In our studies the activities of amylase, cellulase and invertase were found to be significantly higher in organic fields than in conventional field which supports the earlier [24, 25]. This may be attributed to a higher level of carbon sources which accelerate the growth and activity of microorganisms. The declining trend of all selected enzymes with increasing soil depth is an indication of gradual depth dependent reduction of microbial activity. The soil parameters and the microbial activities could be significantly affected by the seasons and soil depths indicating that the activities and density of soil microbes were highly influenced by the soil physicochemistry and depth (Table 3). Improved enzyme activity in winter

Table 3: Analysis of variance of parameters of rice field soil

	Season (S)		Depth (D)		Farming systems (F)		S, D & F		
Variable	C.D	F	C.D	F	C.D	F	C.D	F	C.V (%)
Bacteria	0.73	114717	0.95	1188	0.61	59178	2.32	40.53	0.82
Fungi	1.31	59	1.69	24.8	1.07	128	4.15	3.92	6.36
Amylase	0.39	3331	0.50	48.8	0.32	6195	1.23	17.82	1.07
Invertase	2.28	192	2.95	6.54	1.86	357	7.22	24.79	6.01
Cellulase	0.47	444	0.60	214	0.38	10919	1.48	20.35	1.42
SOC	0.08	328	0.11	91	0.07	11489	0.27	3.42	2.62
MN	0.81	310	1.04	209	0.66	2284	2.54	6.56	1.13
AP	0.63	221	0.81	79	0.51	951	1.99	1.73*	5.58
AK	1.09	2190	1.4	1540	0.88	1975	3.44	39.46	1.69

Note: *Not significant; all other F values were found significant at p = 0.01. Abbreviations: SOC-soil organic carbon; MN-mineralizable nitrogen; AP-available phosphorus; AK- available potassium

Table 4: The coefficients of correlation between depth wise variation of bacterial, fungal density and carbohydrate enzymes' activity of organic (O) and conventional (C) rice field soil

		Amylase		Invertase		Cellulase	
Microbes	Seasons	O	C	O	C	O	C
Bacteria	Winter	0.99	0.96	0.85	0.99	0.93	0.98
	Rainy	0.98	0.79	0.96	0.73	0.89	0.71
	Summer	0.96	0.98	0.95	0.87	0.89	0.97
Fungi	Winter	0.95	0.90	0.83	0.86	0.99	0.81
	Rainy	0.95	0.88	0.95	0.94	0.98	0.77
	Summer	0.94	0.90	0.93	0.95	0.90	0.97

Note: All coefficients were found significant at p = 0.01

corresponds to maximum bacterial and fungal population in this season. In tropical climate the winter season provides moderate temperature and soil moisture conditions which are likely to accelerate microbial population and enzyme activities.

Correlation analysis (Table 4) revealed highly significant relationship between bacterial and fungal population and the enzyme activities irrespective of the farming systems. A highly significant correlation between bacterial population and activities of some other soil enzymes such as phosphatase and urease was reported earlier [26].

In conclusion, it was apparent that organically managed rice field shows superior microbial population and soil enzyme activities in comparison to conventional managed field. It can further be inferred that microbial population and enzyme activity in soil are sensitive to organic inputs and environmental factors which vary during seasons.

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REFERENCES

1. Dick, R.P., 1997. Soil enzyme activities as integrative indicators of soil health. In Biological Indicators of Soil Health, Eds., Pankhurst, Doube, C.E. and B.M. Gupta. CAB International, Wellingford, pp: 121-156.
2. Tabatabai, M.A., 1994. Enzymes, in: Weaver, R.W., S. Augle, P.J. Bottomly, S. Smith, A. Tabatabai and A. Wollum, Eds., Methods of soil analysis. Part 2. Microbial and biochemical properties, Soil Science Society American J., 5: 775-833.
3. Burns, R.G., 1978. History of abiotic soil enzyme responses, Enzym. Academic Press, London, pp: 58-118.
4. Dick, R.P., J.A. Sandor and, N.S. Eash, 1994. Soil enzyme activities after 1500 years of terrace agriculture in the Colea valley, Peru. Agric. Ecosystem Environ., 50: 123-131.
5. Bandick, A.K. and R.P. Dick, 1999. Field management effects on soil enzyme activities. Soil Biology and Biochemistry, 21: 1471-1479.
6. Machulla, G., M.A. Burns and K.M. Scow, 2005. Microbial properties of mine spoil materials in the initial stages of soil development. Soil Science Society of American J., 69: 1069-1077.
7. Krishnakumar, S., A. Saravanan, S.K. Natarajan, V. Veerabadran and S. Mani, 2005. Microbial population and enzymatic activity as influenced by organic farming. Research J. Agriculture and Biological Sci., 1(1): 85-88.
8. Walkley, A. and I.B. Black, 1934. An examination of determining soil organic matter a modification of the chromic acid titration method, Soil Sci., 37: 29-38.
9. Subbaiah, B.V. and H.L. Asija, 1956. A rapid procedure for estimation of available nitrogen in soils. Current Sci., pp: 254-260.
10. Olsen, S.R., C.V. Cole, F.S. Watanabe and L.A. Dean, 1954. Estimation of available phosphorus in soils by extraction with sodium bicarbonate, U.S.D.A. Circular, 939: 1-19.
11. Lu, R.K., 1999. Methods of Soil and Agricultural Chemistry (in Chinese), China Agriculture Science and Technology Press, Beijing, pp: 18638.
12. Johnson, L.F. and E.A. Curl, 1972. Methods for the research on ecology of soil borne plant pathogens, Burgess Publishing Co., Minneapolis, Minnesota,
13. Mishra, P.C., R.K. Mohanty and M.C. Dash, 1979. Enzyme activity in subtropical surface soils under pasture. Indian J. Agri. and Chemistry, 19: 19-24.
14. Gomez, A.A. and K.A. Gomez, 1984. Statistical procedures for agriculture research, John Wiley and Sons, New York, pp: 680.
15. Araujo, A.S.F., L.F.C. Leite, V.B. Santos and R.F.V. Carneiro, 2009. Soil microbial activity in conventional and organic agricultural systems, Sustainability, 1: 268-276.
16. Singh, Y., B. Singh, J.K. Ladha, R.K. Gupta, O.P. Meelu and E. Pasuquin, 2005. Long term effects of organic inputs on yield and soil fertility in the rice wheat rotation. Soil Science Society of American J., 68: 845-853.
17. Shukla, A.K., B.K. Tiwari and R.R. Mishra, 1989. Temporal and depth wise distribution of microbes, enzyme activities and soil respiration in potato field soil under different agricultural systems in Northeastern hill region of India, Rev. Ecol. et de Biol. du Sol., 26: 249-265.
18. Jha, D.K., G.D. Sharma and R.R. Mishra, 1992. Soil microbial population numbers and enzymatic activity in relation to attitudes and forest degradation. Soil Biology and Biochemistry, 24: 761-767.

19. Tangjang, S., S. Deb, A. Arunachalam, V. Melkania, K. Arunachalam, K. Shrivastav and S. Brahma, 2009. Tribal communities and vegetational characteristics in traditional agroforestry systems of Northeast India. Indian J. Agroforestry, 6: 73-80.
20. Classen, A.T., S.T. Overby, S.C. Hart, G.W. Koch and T.G. Whitam, 2007. Season mediates herbivore effects on litter and soil microbial abundance and activity in semi arid woodland. Plant Soil, 295: 217-227.
21. Arunachalam, K.A., R.S.T. Arunachalam and H.N. Pandey, 1997. Dynamics of microbial population during the aggregation phase of a selectively logged tropical humid forest in Northeastern India. Tropical Ecol., 38: 333-341.
22. Dinesh, R., R.P. Dubey, A.N. Ganeshamurthy and P.G. Shyam, 2000. Organic manuring in rice based cropping system: Effects on soil microbial biomass and selected enzyme activities. Current Sci., 79(12): 1716-1719.
23. Gajda, A. and S. Martyniuk, 2005. Microbial biomass C and N and activity of enzymes in soil under winter wheat grown in different crop management systems. Polish J. Environmental Studies, 14: 159-163.
24. Fernandes, S.A.P., B. Bettiol and C.C. Cerri, 2005. Effect of sewage sludge on microbial biomass, basal respiration, metabolic quotient and soil enzymatic activity. Appl. Soil Biol., 30: 65-77.
25. Ge, G.F., Z.J. Li, L.G. Zhang, M.G. Xu, J.B. Zhang, J.K. Wang, X.L. Xie and Y.C. Liang, 2009. Geographical and climatic differences in long term effect of organic and inorganic amendments on soil enzymatic activities and respiration in field experimental stations of China. Ecological Complexity, 6: 421-431.
26. Cheng, H. and Z. Cao, 2007. Size and activity of the soil microbial biomass and soil enzyme activity in long term field experiment, World J. Agri. Sci., 3(1): 63-70.