



# Influence of Soil Ameliorants, Manures and Fertilizers on Bacterial Populations, Enzyme Activities, N Fixation and P Solubilization in Peanut Rhizosphere under Lateritic Soil

M. Basu<sup>1\*</sup>, P. B. S. Bhadoria<sup>1</sup> and S. C. Mahapatra<sup>1</sup>

<sup>1</sup>Indian Institute of Technology, Kharagpur 721 302,  
West Bengal, India

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## ABSTRACT

The aim of this study was to investigate relative efficacy of different organic wastes like farmyard manure (FYM) and water hyacinth (WH) and industrial wastes like paper factory sludge (PFS) on balancing with chemical fertilizers (CF) along with soil ameliorants viz., lime (L) or rice husk ash (RHA), another industrial waste, on dry matter production and biological properties of the rhizosphere soil of peanut (*Arachis hypogaea*), grown as intercrop with sabai grass (*Eulaliopsis binata*) in acid lateritic soil. Population of symbiotic nitrogen fixing and phosphorus solubilizing bacteria, activity of dehydrogenase and phosphatase enzymes (i.e. acid and alkaline phosphomonoesterases), nitrogen accumulation in nodules and phosphorus solubilizing power of rhizosphere soil were measured after 25, 50, 75 and 100 days after sowing (DAS) of peanut for two years. Results showed significant effects of nutrient sources and growth stages of the crop on the microbial activities. Higher values of all the biological properties and plant growth parameters were recorded significantly under the integrated application of CF and any of the organic or industrial wastes over sole application of CF. Among three organic or industrial wastes WH was superior to others regarding microbial activities at 25 DAS, whereas PFS became superior at 50, 75 and 100 DAS. Application of lime or RHA improved the activity of dehydrogenase and alkaline phosphomonoesterase enzymes, while decreased acid phosphomonoesterase activity. This study revealed that integrated application of organic or industrial wastes, soil ameliorants and inorganic fertilizer, could improve the biological properties of an acid lateritic soil as well as the dry matter production of peanut, intercropped with sabai grass under lateritic soil.

**Keywords:** Peanut, Industrial waste, Dehydrogenase, Phosphomonoesterase, N fixation, P solubilization, Acid soil;

## 1. INTRODUCTION

Organic matter is involved in the enhancement of soil quality since it acts on soil structure, nutrient storage and biological activity. Soil microorganisms are significant determinants of organic matter

\* Corresponding author: E-mail: manishabckv@yahoo.co.in

decomposition, soil nutrient status, crop health, and overall crop productivity. The extent of soil organic matter turnover is mainly controlled by the size and activity of the microbial biomass. For this reason, soil biological and biochemical parameters may have a role as early and sensitive indicators of soil ecological stress and restoration (Izquierdo et al., 2003). The native or added organic matter transformations in the soil involve many reactions which are catalyzed by enzymes existing outside the microorganisms and plant root system (Sarapatka, 2003). The presence and activity of enzymes is vital for all biochemical transformations in soil, thus the study of soil enzymatic activities provides insight into microbial dynamics and populations (Riffaldi et al., 2002). Knowledge of the spectrum of enzymatic activities of a soil is important since it indicates the potential of the soil to permit the basic biochemical processes necessary for maintaining soil fertility. Enzymatic activities in relation to the cycling of nitrogen (ammonification, nitrification, denitrification) or phosphorus (release of inorganic phosphorus) in soil have been used to evaluate the fertility of the soil (Aon and Colaneri, 2001; Brohon et al., 2001). Dehydrogenase activity is assayed as an estimation of overall microbial activity. Phosphatases are often measured because of their importance in the phosphorus cycles (Aon and Colaneri, 2001).

Data on the magnitude of changes in soil microbial activities after application of soil ameliorants and different organic and inorganic nutrient sources in legume rhizosphere under grass-legume intercropping system of subtropical agro-ecosystems is very scanty. The objective of this study was to determine the effects of soil ameliorants and different sources of nutrients on the biological properties of rhizosphere soil of peanut, intercropped with sabai grass in the lateritic sub-tropics of eastern India.

Sabai grass (*Eulaliopsis binata* (Retz.) C.E. Hubb), a perennial plant with about 54.5% of cellulosic material and high quality fiber, is used as an excellent raw material in paper pulp industries and in agro-based rural industries for making ropes and other rope based utility items (Mohapatra et al., 2001), for which sabai grass has an important role in tribal economics of many regions of India (Anonymous, 2002a; Basu et al., 2006) as well as many Asian countries like China, Pakistan, Nepal, Bhutan, Myanmar, Thailand, Malaysia and Philippines (Yong, 1994; Basu et al., 2006). Recently understanding the structure, mechanical and thermal behavior of sabai grass fibre has opened up new avenues for the utilization of this fibre (Chand and Rohatgi, 1992). It can also be used as a filler material in plastics and in mud matrix after suitable pretreatment to the surface. Wide spacing and initial slow growth rate of sabai grass provides ample scope for intercropping in association with legumes in the initial 1-2 years (Mohapatra et al., 2001). Intercropping of various legumes like greengram or cowpea or blackgram in the initial 2-3 years of establishment with sabai grass proved superior over the sole crop of sabai grass both in total productivity, crude fiber content and net return (Barik, 2002). Being the most important oilseed crop of India, peanut has been cultivated as an intercrop with sabai grass for initial two years in the present investigation.

## **2. MATERIAL AND METHODS**

### **2.1 EXPERIMENTAL SITE**

The experiment was carried out in the research farm, situated in the lateritic belt of south-western region of West Bengal, India. The site is intersected by 22°19' North latitude and 87°19' East longitude at a distance of 115 km from the Bay of Bengal and has an elevation of 44.0 m above the mean sea level. The climate of the region is warm and humid. The average annual rainfall is 1400 mm, about 80% of which is received from mid June to mid October.

### **2.2 SOIL**

The soil is acid lateritic (Haplustalf) having sandy loam texture with 61% sand, 21% silt and 18% clay. The soil is shallow and coarse textured which results in low plant available water capacity

causing moisture stress during crop growing period. Intensive leaching causes nutrient losses and release of free Fe and Al causing toxicity and nutrient imbalances in terms of N, P, K and Zn. The acidification of this soil with pH less than 5.5 causes P fixation and reduces the availability of K, Ca and Mg. It is low in organic carbon and N content. The detailed physical, chemical and biological properties of the experimental soil are presented in Table 1.

**Table 1: Physical, chemical and biological properties of experimental soil (0-20 cm)**

Particulars	Value	Methods
Sand (%)	61.3	Pipette Method (Piper, 1966)
Silt (%)	21.4	
Clay (%)	17.3	
Bulk density, Mg m <sup>-3</sup>	1.64	Core Sampler (Piper, 1966)
pH (1 : 2.5:: Soil : Water)	5.20	Glass Electrode pH Meter (Jackson, 1973)
Organic carbon (g kg <sup>-1</sup> )	2.9	Glass Electrode pH Meter (Jackson, 1973)
Total N, %	0.049	Modified Kjeldahl Method (Chapman and Pratt, 1961)
Available N, mg kg <sup>-1</sup>	65.60	Alkaline KMnO <sub>4</sub> (Subbaiah and Asija, 1956)
Total P, %	0.039	Perchloric Acid Method (Jackson, 1973)
Available P, mg kg <sup>-1</sup>	5.12	NH <sub>4</sub> F Extraction (Jackson, 1973)
Total K, %	0.058	HF acid Decomposition (Chapman and Pratt, 1961)
Available K, mg kg <sup>-1</sup>	44.53	NH <sub>4</sub> OAc Extraction (Jackson, 1973)
Population of P solubilizing bacteria (log no. CFU g <sup>-1</sup> soil)	4.62	Pikovskaia's method (Pikovskaia, 1948)
Dehydrogenase enzyme activity (µg TPF g <sup>-1</sup> soil h <sup>-1</sup> )	6.99	TTC method (Thalman, 1968)
Acid phosphatase enzyme activity (µg PNP g <sup>-1</sup> soil h <sup>-1</sup> )	178.3	Tabatabai and Bremner, 1969; Eivazi and Tabatabai, 1977
Alkaline phosphatase enzyme activity (µg PNP g <sup>-1</sup> soil h <sup>-1</sup> )	41.2	Do
P solubilizing power (mg insoluble P solubilized 15 mg <sup>-1</sup> insoluble P g <sup>-1</sup> soil)	0.045	Pikovskaia's method (Pikovskaia, 1948)

\*Moisture content on oven dry weight basis

## 2.3 TREATMENTS

The experiment was conducted for three years during 2003-'04, 2004-'05 and 2005-'06. A uniform level of nutrients @ 50 kg N, 25 kg P and 25 kg K ha<sup>-1</sup> was maintained through chemical fertilizers (CF) alone and in combination with organic or industrial wastes. Organic wastes viz., farmyard manure (FYM) and water hyacinth (WH) and industrial waste viz., paper factory sludge (PFS) were applied at 50% of recommended N level (i.e., 25 kg ha<sup>-1</sup>) as organic sources of nutrients. The balance 25 kg ha<sup>-1</sup> of N and the difference in P and K from the applied levels of different organic nutrient sources were met through CF. The treatments comprising organic/industrial wastes + CF (FYM+CF, WH+CF and PFS+CF) were tested alone as well as in combination with soil ameliorants viz., RHA @ 5 t ha<sup>-1</sup> or lime @ 2 t ha<sup>-1</sup>. N, P and K added through RHA were not taken into consideration while maintaining the recommended NPK level. An absolute control was also included

for treatment comparison. All together 11 treatment combinations (i.e., CF, FYM+CF, FYM+L+CF, FYM+RHA+CF, WH+CF, WH+L+CF, WH+RHA+CF, PFS+CF, PFS+L+CF, PFS+RHA+CF and Control) with three replications were laid out. The experiment was repeated for consecutive three years with the application of similar organic and inorganic nutrient sources each year. Physical and chemical properties of organic and industrial wastes used in the experiments have been given in Table 2. The effect of these treatments was studied on sabai grass-peanut intercropping system during 2003 and 2004 and on sole sabai grass during 2005. Peanut was grown with sabai grass as an intercrop at 1:2 ratio (two rows of peanut after every row of sabai grass) in the initial two years, since smothering effect due to vigorous growth of sabai grass did not allow growing peanut in the subsequent year. In this paper the effect of nutrient sources on the soil biological properties in the peanut rhizosphere has been presented. Total quantity of P and K and half of N was applied as basal at the time of sowing/planting/regrowth of crops during rainy season of each year. Remaining half N was top dressed in equal split through side dressing to sabai grass at 30 and 60 days after planting or regrowth. Urea, single super phosphate and muriate of potash were used as the sources of N, P and K respectively.

**Table 2: Physical and chemical properties of organic and industrial wastes used in the experiments**

Particulars	Organic wastes		Industrial wastes	
	Farmyard manure	Water hyacinth	Paper factory sludge	Rice husk ash
Colour	Brownish black	Brownish black	Blackish ash	Blackish ash
Basic organic material	Crop wastes and cow dung	Whole plant water hyacinth	Wastes of paper factory	Rice husk
Texture	Small lumps	Dried leafy	Coarse layery dust	Coarse dust
Bulk density, Mg m <sup>-3</sup> *	0.48	0.43	0.58	0.43
pH**	5.54	5.35	5.88	7.57
Organic carbon, %	20.9	21.5	25.2	4.89
N, %	0.83	1.23	0.71	0.06
P, %	0.26	0.39	0.15	0.31
K, %	0.65	2.09	0.30	0.14
C:N	25:1	17:1	36:1	82:1

\*Oven dry weight basis; \*\*Material : Water = 1:5

Since sabai grass is perennial in nature 10-12 slips per hill were planted once during rainy season of 2003 (July) at a spacing of 100 cm x 50 cm for row-to-row and plant-to-plant respectively. In case of peanut, seeds were treated with the fungicide Bavistein @ 3 g kg<sup>-1</sup> of seed then sown manually by putting 2-3 seed per hole in rows with spacing of 30 cm x 10 cm for row-to-row and plant-to-plant respectively. Thinning of seedlings was done between 7 to 10 days after sowing (DAS). Earthing up was done at 30 and 60 DAS with 'kharpi' (small hand spade). Data on dry matter production was recorded at 25, 50, 75 and 100 DAS from randomly selected 10 plants.

## 2.4 SOIL SAMPLING

Rhizosphere soils were collected at different growth stages of peanut (25, 50, 75 and 100 DAS), by uprooting four plants from each plot and keeping the soil around root system intact. After removing the bits of plant roots and other debris, the soil strongly adhered to the roots was immediately used without drying for determination of soil biological properties. The population of symbiotic N fixing

bacteria and P solubilizing bacteria, activity of dehydrogenase, acid phosphatase and alkaline phosphatase enzymes, N accumulation in nodules and P solubilizing power were determined.

## 2.5 CHEMICAL ANALYSIS OF SOIL SAMPLE, PLANT SAMPLE AND ORGANIC AND INDUSTRIAL WASTES

Before starting of the experiment soil samples from each plot were collected randomly up to a depth of 20 cm by soil auger and representative samples were prepared and stored for chemical analysis by following standard procedures (Jackson, 1973). Soil physical properties like soil texture and bulk density, and chemical properties such as pH, organic carbon, concentration of N, P and K were analyzed as per the standard procedures as mentioned in Table 1.

The N and P content of FYM, WH, PFS and peanut haulm were estimated by following the methods of Chapman and Pratt (1961), while K content was determined by wet digestion method as described by Jackson (1973). N, P and K concentrations of RHA were determined by the procedures followed in soil analysis (Table 1).

## 2.6 VIABLE COUNT OF BACTERIA

Agar plates with appropriate media following serial dilution technique and pour plate method were used for enumeration of bacterial population (Pramer and Schmidt, 1965). The media used were yeast extract mannitol agar (YEMA) medium for symbiotic N fixing bacteria (Vincent, 1970) and Pikovskaia's agar medium for P solubilizing bacteria (Pikovskaia, 1948).

## 2.7 DEHYDROGENASE ACTIVITY

The method is based on the estimation of the triphenyl tetrazolium chloride (TTC) reduction rate to triphenyl formazan (TPF) during composting after incubation at  $30 \pm 1$  °C for 24 h. All procedures were performed under diffused light because of the light sensitivity of TTC and TPF. The dehydrogenase activity was determined with the following formula.

$$\text{Dehydrogenase activity } (\mu\text{g TPF g}^{-1}\text{dwt}) = \frac{\text{TPF } (\mu\text{g ml}^{-1}) \times 50}{\text{dwt} \times W}$$

Where, TPF ( $\mu\text{g ml}^{-1}$ ) = found from standard curve;  
dwt = Dry weight of 1 g soil;  
W = Weight of the moist soil;  
50 = Total volume of the solution added to the soil.

## 2.8 PHOSPHATASE ACTIVITY

The method is based on the determination of *p*-nitrophenol (PNP) released after the incubation of the soil with *p*-nitrophenol phosphate for 1 h at  $37 \pm 1$  °C using modified universal buffer, MUB (pH 6.5 for the assay of acid phosphatase, pH 11.0 for the assay of alkaline phosphatase) as substrate.. The phosphatase activity was determined with the following formula.

$$\text{Acid or alkaline phosphatase activity } (\mu\text{g } p\text{-nitrophenol g}^{-1}\text{dwt h}^{-1}) = \frac{C \times V}{\text{Dwt} \times C_w \times t}$$

Where, C = Measured concentration of *p*-nitrophenol ( $\mu\text{g ml}^{-1}$  filtrate) after correcting the result for the control;  
V = Total volume of the suspension (ml);  
Dwt = Dry weight of 1 g moist soil;  
Cw = Weight of the soil sample used (g); t = Incubation time (h).

## 2.9 PHOSPHORUS SOLUBILIZING POWER

The phosphate solubilizing power of the above soils was determined by estimating soluble phosphorus after incubating 1 g soil in culture tubes for 15 days, at  $30 \pm 1$  °C in 15 ml Pikovskaia's broth (Pikovskaia's medium, without agar), containing insoluble phosphate, followed by estimation of soluble phosphorus in the broth.

## 2.10 STATISTICAL ANALYSIS

The recorded data were analyzed statistically following the standard procedure as described by Gomez and Gomez (1984). Correlation and regression coefficients of various parameters were calculated. The means were compared using least significant difference (LSD) test, with a significance level of  $P \leq 0.05$ . Analyses of variance (two way factorial ANOVA) were carried out using a randomized complete block design (nutrient sources x plant growth stages).

## 3. RESULTS AND DISCUSSION

### 3.1 BACTERIAL POPULATION

The number of symbiotic N fixing bacteria and P solubilizing bacteria in the peanut rhizosphere soil at different growth stages (25, 50, 75 and 100 DAS) were significantly ( $P \leq 0.05$ ) higher under different nutrient sources as compared to control (Table 3).

**Table 3: Population of bacteria in the rhizosphere soil of peanut at different growth stages as influenced by different nutrient sources during 2003 and 2004**

Nutrient sources	Bacterial population (Log no. CFU g <sup>-1</sup> soil)			
	Symbiotic N fixing bacteria		P solubilizing bacteria	
	2003	2004	2003	2004
CF	5.30	4.86	5.06	4.66
FYM+CF	6.46	5.73	6.01	5.46
FYM+L+CF	7.05	6.35	6.62	6.10
FYM+RHA+CF	7.45	6.74	7.01	6.48
WH+CF	6.47	5.69	5.92	5.41
WH+L+CF	7.08	6.24	6.52	5.99
WH+RHA+CF	7.40	6.65	6.88	6.36
PFS+CF	6.51	5.79	6.06	5.48
PFS+L+CF	7.15	6.45	6.73	6.20
PFS+RHA+CF	7.56	6.82	7.14	6.60
Control	4.76	4.30	4.58	4.15
<b>S.E(m)±</b>	<b>0.116</b>	<b>0.076</b>	<b>0.118</b>	<b>0.059</b>
<b>LSD (P=0.05)</b>	<b>0.29</b>	<b>0.19</b>	<b>0.29</b>	<b>0.15</b>
Growth stages, Days after sowing				
25	6.41	5.72	6.03	5.57
50	7.13	6.41	6.68	6.13
75	6.85	6.15	6.46	5.89
100	6.23	5.58	5.76	5.29
<b>S.E(m)±</b>	<b>0.070</b>	<b>0.046</b>	<b>0.071</b>	<b>0.036</b>
<b>LSD (P=0.05)</b>	<b>0.17</b>	<b>0.11</b>	<b>0.18</b>	<b>0.09</b>

CF = Chemical fertilizer; FYM = Farmyard manure; WH = Water hyacinth; PFS = Paper factory sludge; L = Lime; RHA = Rice husk ash; CFU = Colony forming units;

In all the treatments the bacterial population attained its maximum value at 50 DAS and thereafter decreased significantly ( $P \leq 0.05$ ) till 100 DAS. Among three organic sources, WH based treatment combinations resulted maximum proliferation of bacteria at initial growth stage and there was significant difference with PFS based treatments. However, from 50 DAS onwards PFS based treatments recorded superior results as compared to WH or FYM. This might be due to the fact that WH with comparatively lower C:N ratio (17:1) (Table 2) decomposed quickly and thereby promoted higher bacterial growth at initial stage, whereas PFS with maximum C:N (36:1) (Table 2) ratio resulted an initial immobilization effect (Burgos et al., 2006). Farmyard manure with intermediate C:N ratio (25:1) (Table 2) showed moderate effect. Combined application of any of these organic sources along with CF significantly improved the bacterial population over CF alone in all the growth stages, which is in agreement with Sivapalan et al. (1993) and Lee et al. (2004), who also found remarkably higher population of microbes in organic manure applied area than in the conventional area.

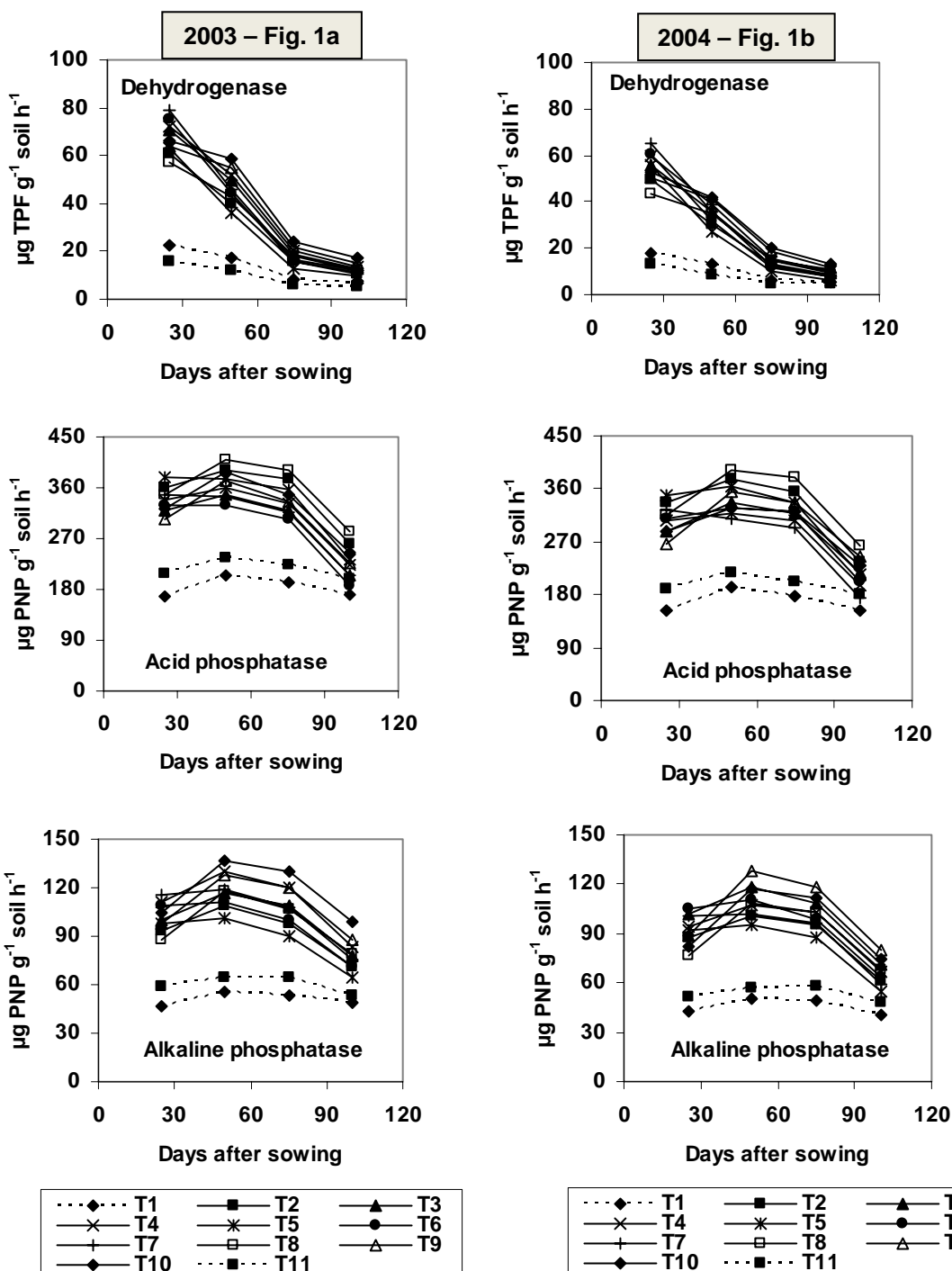
Application of soil ameliorants like lime and RHA further increased the bacterial population than similar treatment combinations but without lime or RHA. The beneficial effect of soil ameliorants was due to rise in the soil reaction up to 6.54 (unpublished data of this experiment) with industrial/organic wastes + CF from an initial value of 5.20 (Table 1) and it was favorable for the growth of bacteria. Greater proliferation of *Bradyrhizobium* population with increasing pH towards neutral was also reported by Fettel et al. (1998). The enhancement of P solubilizing bacteria due to liming was also reported by Barroti and Nahas (2000). Between lime and RHA, RHA was more promising in improving the bacterial population as compared to lime. The beneficial effect of RHA on viability and nodule forming ability of *Rhizobium sp.* was reported by Deka and Baruah (1992).

### 3.2 ACTIVITY OF ENZYMES

In general irrespective of treatment variations, the activity of dehydrogenase enzyme (DH) in the rhizosphere soil of peanut was found to be maximum at initial growth stage and thereafter decreased till 100 DAS (Figures 1a & 1b). Among different nutrient sources, WH based treatments showed faster rate of decrement in DH activity from 25 up to 75 DAS, while PFS based treatments recorded steady decrease up to 50 DAS, thereafter declines at a faster rate up to 75 DAS. That might be presumably due to the exhaustion of the readily decomposable carbonaceous materials from WH during initial stage, whereas, resistant carbonaceous materials of wide C:N ratio material (PFS) was gradually decomposed by the action of responsible microorganisms in soil probably by the proto-cooperation with N fixers and the cellulose decomposers, that converted cellulose to simple sugars (Mukherjee et al., 1999).

The activity of acid phosphatase (ACP) and alkaline phosphatase (ALP) enzymes significantly ( $P \leq 0.05$ ) increased from 25 to 50 DAS and then decreased at a slow rate up to 75 DAS and at fast rate till 100 DAS. At the initial stage, the rate of decomposition of WH was faster than FYM or PFS and this differential rate of decomposition could be attributed to lower C:N ratio of WH to FYM or PFS, which substantiated the findings of Mukherjee et al. (1990), that N rich materials are metabolized very rapidly. Though the C:N ratio of FYM and WH was close, but FYM being a stabilized organic material decomposed slowly than WH (Mukherjee et al., 1999). Between PFS and FYM, the former one was superior over the later one in all the growth stages except 25 DAS.

The application of CF significantly ( $P \leq 0.05$ ) increased the DH activity, while significantly decreased the activity of phosphomonoesterases (ACP and ALP) as compared to control, although the proliferation of P solubilizing bacteria was significantly higher under CF treated plots than control, as we observed in the earlier section. This is similar with the finding of Criquet et al. (2004) and Ne'ble (2005), who reported that phosphomonoesterases' activity was not correlated to any specific bacterial group, because these enzymes are produced by a large fraction of the soil microflora (Hysek and Sarapatka, 1998).



T1 = Chemical fertilizer (CF); T2 = Farmyard manure (FYM)+CF; T3 = FYM+Lime (L)+CF; T4 = FYM+Rice husk ash (RHA)+CF; T5 = Water hyacinth (WH)+CF; T6 = WH+L+CF; T7 = WH+RHA+CF; T8 = Paper factory sludge (PFS)+CF; T9 = PFS+L+CF; T10 = PFS+RHA+CF; T11 = Control

**Fig. 1a & 1b: Activity of enzymes in the rhizosphere soil of peanut as influenced by different nutrient sources during 2003 (1a) & 2004 (1b)**



In addition, investigation of fungal community was not performed in the present study. Monitoring the fungal community could be important because acid phosphomonoesterases in soil are also produced by plant roots and fungi whereas alkaline phosphomonoesterases are produced by bacteria (Tabatabai, 1994). Several studies also showed negative correlations between available P and phosphatase activities (Lima et al., 1996; Olander and Vitousek, 2000; Moscatelli et al., 2005). This phenomenon could also be explained by a competitive inhibition of phosphatases by phosphate ions or by a negative-feed back of phosphate ions on PHO genes resulting in a repression of phosphatase synthesis by microorganisms (Oshima et al., 1996; Criquet et al., 2007).

The treatments comprising CF along with any of the organic sources considerably increased the activity of these three enzymes as compared to sole CF. In an experiment, Lee et al. (2004) also stated that the enzyme activity in organic amended soil increased by an average 2-4 folds as compared with the unamended soil. These results were similar to our finding that DH activity in the rhizosphere soil of organic manure treated plots was about 1.5-5 times higher than that of control or sole CF treatment. The increase in DH, ACP and ALP activity after additions of organic or industrial wastes viz., FYM or WH or PFS along with CF to soils had been generally attributed to the fact that enzyme activities directly associated to organic matter and to microbial response to soluble sugars of the added materials (Nannipieri et al., 1983; Hojati and Nourbakhsh, 2006). Lalande et al. (2003) also reported improvement in phosphatase activity due to application of paper mill sludge and thereby corroborated our results.

There was remarkable variation in the activities of DH, ACP and ALP due to application of soil ameliorants. Both lime and RHA significantly promoted the activity of DH and ALP, but reduced the activity of ACP, when applied along with organic/industrial wastes + CF. The RHA based treatments showed higher DH, ACP and ALP activity than lime based treatments. Improvement of soil pH due to application of lime or RHA enhanced the activity of DH enzyme significantly ( $P \leq 0.05$ ) and such increment was to the extent of 36% and 49% respectively as compared to similar combinations but without soil ameliorants. The enhanced DH activity after lime addition was probably due to the increase in microbial population as reported by Ray (1985). This is also apparent from a significant ( $P \leq 0.01$ ) and positive correlation between bacterial population and DH activity (Table 5).

The activity of ACP was declined to the extent of 15% due to application of lime or RHA, while the ALP activity was improved by about 9% (average of two years). The reason might be due to rise of soil pH to nearly neutral range (6.54, unpublished data of this investigation) after application of lime or RHA, since the acid phosphomonoesterase activity dominates in acidic soils whereas the alkaline phosphomonoesterase activity dominates in neutral and alkaline soils (Dick and Tabatabai, 1992; Dick et al., 2000; George et al., 2002). The different responses of ACP and ALP activity to RHA and lime were in consonance to the previous findings that phosphatases being inducible enzymes, the intensity of their excretion by plant roots and microorganisms was determined by their requirement for orthophosphate, which was affected by soil pH (Acosta-Martinez and Tabatabai, 2000).

### **3.3 NITROGEN CONCENTRATION OF NODULES AND PHOSPHORUS SOLUBILIZING POWER OF THE RHIZOSPHERE SOIL**

The amount of N concentration of nodules and insoluble P solubilized  $g^{-1}$  of rhizosphere soil of peanut at different growth stages are presented in Table 4. Irrespective of treatment variations, maximum amount of N accumulation in the nodules and P solubilizing capacity of the rhizosphere soil were observed at 50 DAS. Integrated application of FYM or WH or PFS and CF significantly ( $P \leq 0.05$ ) improved the biological processes like N fixation and P solubilization over CF alone in the order PFS>FYM>WH, although at the initial growth stages of the crops the trend was WH>FYM>PFS. There was no significant difference between FYM and PFS. Higher concentration of water soluble sugar and carbohydrates in the succulent WH over FYM or PFS made it easily decomposable by the microorganisms, which led to higher bacterial proliferation, enzyme activity and these were probably responsible for higher N fixation and P solubilization as compared to FYM and PFS. Positive and significant correlations among bacterial population, enzyme activities and these biochemical parameters further confirmed these results. Increase in P solubilizing power of bacteria due to the application of organic manure was also reported by Saha et al. (1995). Low P solubilizing capacity might be attributed to less enzymatic activity under sole CF application, since only phosphatase enzymes produced by plants and/or microorganisms are able to

hydrolyze organic P into phosphates through hydrolysis of both esters and anhydrides of  $H_3PO_4$  (Criquet et al., 2007). Among these enzymes, acid and alkaline phosphomonoesterases and phosphodiesterases are considered as the predominant phosphatases in most types of soil and litter (Criquet et al., 2004).

**Table 4: N concentration of nodules and P solubilizing power of the rhizosphere soil of peanut at different growth stages as influenced by different nutrient sources during 2003 and 2004**

Nutrient sources	N concentration of nodules (%)		P solubilizing power (mg P solubilized 15 mg <sup>-1</sup> insoluble P g <sup>-1</sup> soil)	
	2003	2004	2003	2004
CF	6.12	5.63	0.215	0.191
FYM+CF	6.78	6.33	0.244	0.217
FYM+L+CF	7.34	6.92	0.273	0.238
FYM+RHA+CF	7.69	7.31	0.283	0.250
WH+CF	6.69	6.26	0.244	0.213
WH+L+CF	7.24	6.86	0.265	0.236
WH+RHA+CF	7.56	7.16	0.277	0.248
PFS+CF	6.76	6.38	0.248	0.220
PFS+L+CF	7.47	7.07	0.281	0.241
PFS+RHA+CF	7.79	7.43	0.290	0.254
Control	4.95	4.57	0.171	0.153
<b>S.E(m)±</b>	<b>0.094</b>	<b>0.161</b>	<b>0.0090</b>	<b>0.0056</b>
<b>LSD (P=0.05)</b>	<b>0.23</b>	<b>0.40</b>	<b>0.022</b>	<b>0.014</b>
<b>Growth stages, Days after sowing</b>				
25	6.71	6.28	0.250	0.221
50	7.68	7.07	0.277	0.245
75	7.08	6.75	0.259	0.232
100	6.32	6.06	0.228	0.198
<b>S.E(m)±</b>	<b>0.057</b>	<b>0.097</b>	<b>0.0054</b>	<b>0.0034</b>
<b>LSD (P=0.05)</b>	<b>0.14</b>	<b>0.24</b>	<b>0.014</b>	<b>0.008</b>

CF = Chemical fertilizer; FYM = Farmyard manure; WH = Water hyacinth; PFS = Paper factory sludge; L = Lime; RHA = Rice husk ash

Lime and RHA based treatments improved these biological activities significantly ( $P \leq 0.05$ ) over sole CF in a comparable manner. Application of lime or RHA along with organic/industrial wastes + CF significantly stimulated biological reactions over similar combinations but without lime or RHA. This might be due to higher bacterial population and enzyme activities under the integrated plant nutrient management systems as compared to only CF. A positive and significant correlation between population of aerobic symbiotic N fixing bacteria and N fixing capacity or between population of P solubilizing bacteria and P solubilizing power was reported by Saha et al. (1995). In our study also, positive and significant ( $P \leq 0.01$ ) correlations among these parameters were observed as presented in Table 5. Raychaudhury et al. (2003) also reported improved N fixation by peanut due to liming and this might be ascribed to the fact that nodules on the lower part of the root system were more efficient to fix greater amount of N than crown nodules throughout the growing season and they might contribute most of the N fixed by the legume plant (Hardarson et al., 1989). Liming increased the root length and lateral root distribution because of increase in exchangeable  $Ca^{2+}$  and  $Mg^{2+}$  levels in the soil and improved the soil structure providing better aeration in the rhizosphere of peanut, which was helpful in better nodulation (Raychaudhury, 2003).

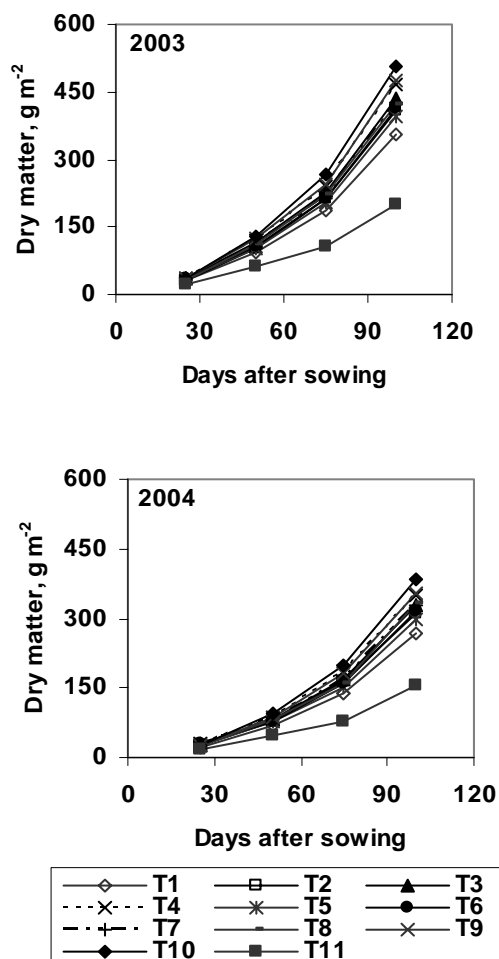
### 3.4 DRY MATTER PRODUCTION AND SHOOT NUTRIENT CONCENTRATION (N, P AND K)

All the treatment combinations containing either FYM or WH or PFS significantly ( $P \leq 0.05$ ) influenced the dry matter production at different growth stages and concentrations of N, P and K in shoot or haulm of peanut at maturity when applied with CF as compared to sole CF or control (Figures 2 and 3).

**Table 5: Linear relationships (*r*-values) among different soil biological parameters of peanut rhizosphere studied for evaluating the effect of different nutrient sources**

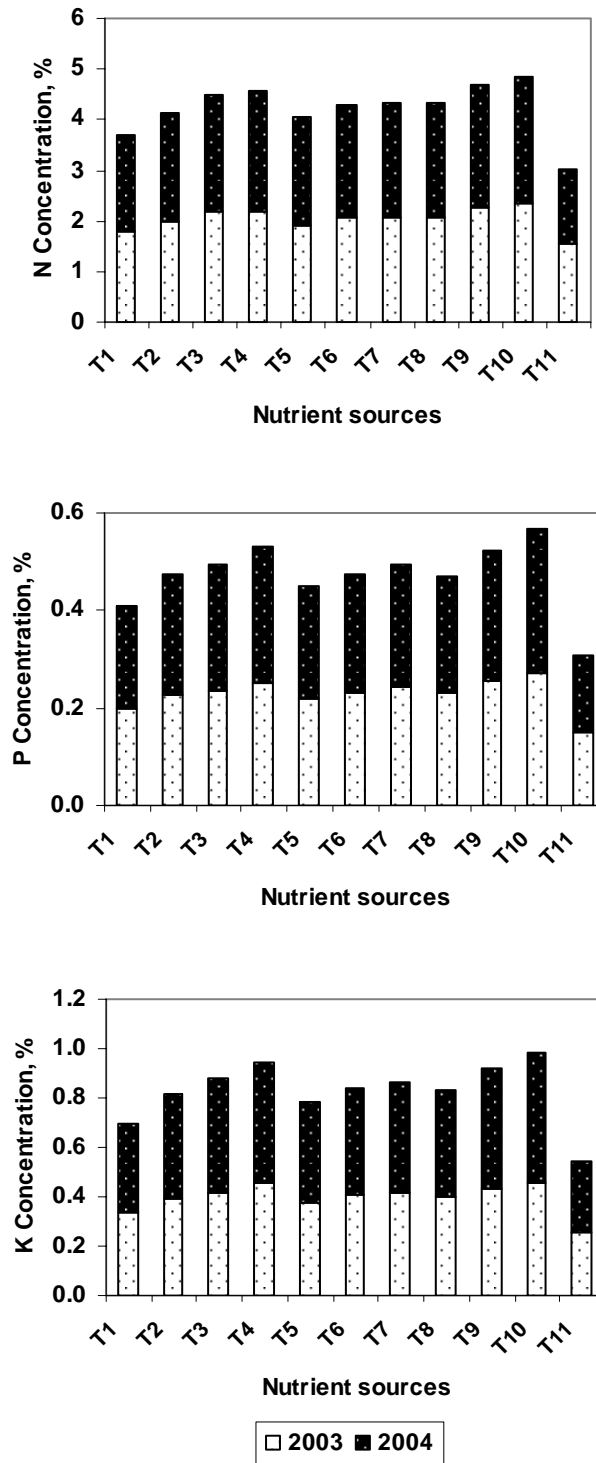
Parameters	Symbiotic N fixing bacteria	P solubilizing bacteria	DH activity	ACP activity	ALP activity	Nodule N Conc.	P solubilizing power	Dry matter production
Symbiotic N fixing bacteria			0.42**			0.92**		0.66**
P solubilizing bacteria			0.45**	0.62**	0.82**		0.83**	0.72**
Dehydrogenase activity						0.52**	0.49**	0.81**
ACP activity							0.64**	0.59**
ALP activity							0.82**	0.78**
Nodule N concentration								0.73**
P solubilizing power								0.79**

\*\* Significant at 1% level; DH= Dehydrogenase; ACP = Acid phosphatase; ALP = Alkaline phosphatase.



T1 = Chemical fertilizer (CF); T2 = Farmyard manure (FYM)+CF; T3 = FYM+Lime (L)+CF; T4 = FYM+Rice husk ash (RHA)+CF; T5 = Water hyacinth (WH)+CF; T6 = WH+L+CF; T7 = WH+RHA+CF; T8 = Paper factory sludge (PFS)+CF; T9 = PFS+L+CF; T10 = PFS+RHA+CF; T11 = Control

**Fig. 2: Dry matter production of peanut at different growth stages as influenced by different nutrient sources during 2003 and 2004**

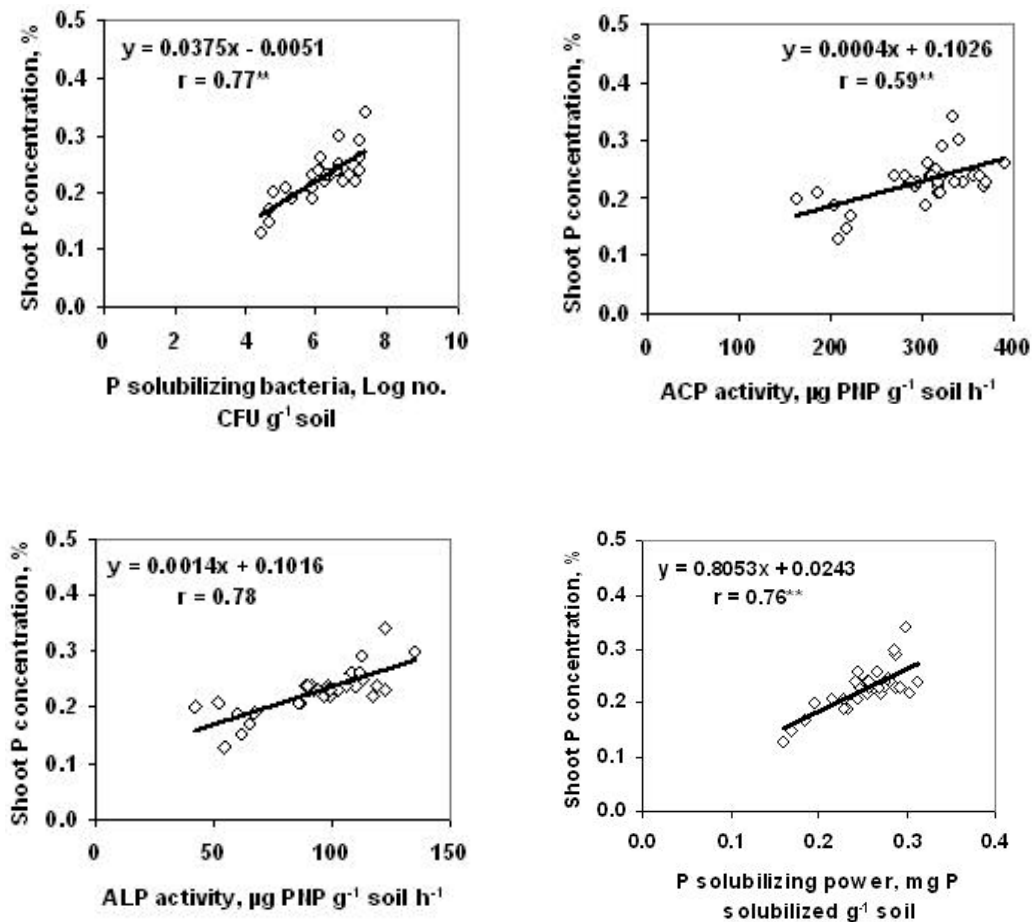


T1 = Chemical fertilizer (CF); T2 = Farmyard manure (FYM)+CF; T3 = FYM+Lime (L)+CF; T4 = FYM+Rice husk ash (RHA)+CF; T5 = Water hyacinth (WH)+CF; T6 = WH+L+CF; T7 = WH+RHA+CF; T8 = Paper factory sludge (PFS)+CF; T9 = PFS+L+CF; T10 = PFS+RHA+CF; T11 = Control

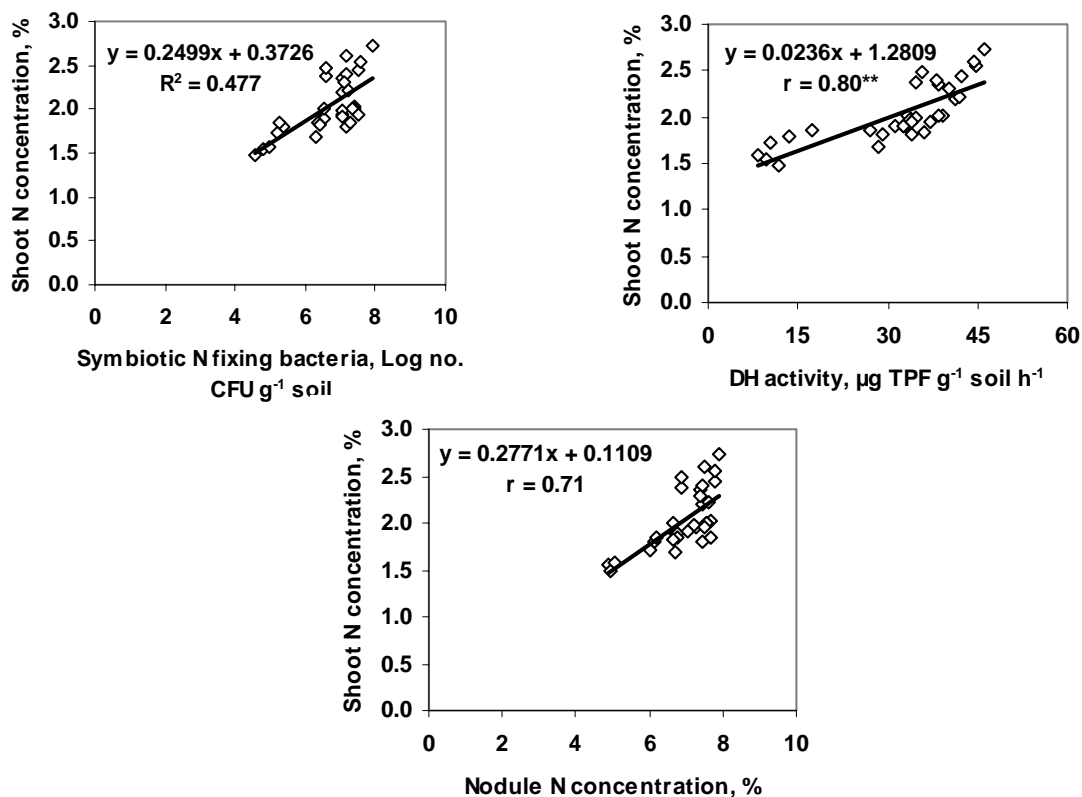
**Fig. 3: Nitrogen, phosphorus and potassium concentration of peanut shoot at harvest as influenced by different nutrient sources during 2003 and 2004**

The concentrations of these macronutrients at maturity were noted to be maximum under fertilization treatments of PFS, soil amendments and CF, which was followed by FYM and then WH under similar combinations.

The treatments where organic sources were combined with soil amendments and CF recorded remarkably higher dry matter production and nutrient concentration than in similar combinations but without soil amendments. Higher shoot N and P concentrations under integrated plant nutrient management systems might be due to higher proliferation of symbiotic N fixing bacteria and P solubilizing bacteria, greater activities of dehydrogenase and phosphomonoesterase enzymes, which led to more concentration of N in the nodules and P solubilizing capacity resulting in enhanced availability of N and P to the plant. Higher availability of nutrients resulted in more nutrient uptakes and dry matter production by the plant. A positive and significant correlation among different biological parameters, dry matter production and shoot N and P concentrations supported these results (Figures 4 and 5).



**Fig. 4: Linear relationships ( $r$ -values) among different biological parameters of rhizosphere soil and shoot N concentration of peanut studied for evaluating the effect of different nutrient sources (\*\* Significant at 1% level;)**



**Fig. 5: Linear relationships ( $r$ -values) among different biological parameters of rhizosphere soil and shoot P concentration of peanut studied for evaluating the effect of different nutrient sources (\*\* Significant at 1% level;)**

#### 4. CONCLUSIONS

Results of the present investigation revealed that soil biological parameters were controlled by short term application of manures and fertilizers along with soil ameliorants and crop development stages of peanut crop. Three of the major contributions of integrated nutrient management system comprising inorganic fertilizers, organic or industrial wastes and soil ameliorants, to the acid lateritic soils were the increased bacterial population, enzyme activities and microbial activities, which are key factors of nutrient availability to the plant. This could be understood from increased shoot nutrient concentration of peanut crop, which led to higher dry matter production. Rich microbial populations and enzyme contents of the organic nutrient sources might have been responsible for improvement of soil biological quality in the peanut rhizosphere. Application of inorganic fertilizers alone although improved the bacterial population, but could not improve phosphomonoesterase activities. However, this had no negative effect on other microbial activities or performance of peanut plant. Therefore it indicated the fact that microbial densities could not be used as the constant indicator of soil biological quality.

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