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Research Article

BIOCHEMICAL CHANGES IN CONTINUOUS APPLICATION OF MANURES AND FERTILIZER UNDER RICE MONOCULTURE OVER FOUR DECADES IN TYPIC HAPLUSTALF

RAMYA C., GOKILA B., BAKIYATHU SALIHA B. AND BASKAR K.
Tamil Nadu Agricultural University, Coimbatore, 641 003, Tamil Nadu, India
*Corresponding Author: Email - singh_gokila@rediffmail.com

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Abstract- Biochemical reactions are the important nutrient transformation processes in organic and inorganic substance in soil environment through the catalytic activity of biomolecules called enzymes. Long term manures and fertilizer applications and continuous cropping of rice monoculture have a significant impact on soil enzymatic activities in a soil system. The investigation assessed the effect of long term manure and fertilizer usage on soil enzyme activities through organic manures *viz*., FYM, GLM and UC @ 12.5 t ha^{-1} and omission of N, P, K and addition of NPK with recommended dose of 150:50:50 kg N, P_2O_5 and K_2O respectively. Combined application of manures GLM @ 12.5 t ha^{-1} along with NPK application was significantly increased the urease (20.61 %), phosphatase (15.21%) and dehydrogenase (24.17%) over control in tillering stage of rice monoculture. Regarding soil available nutrients in post harvest soil, application of GLM @ 12.5 t ha^{-1} along with NPK fertilizers increased the soil available $KMnO_4$–N (10%), Olsen-P (43.31%) and NNH_4OAc-K (35.97%) over control. Continuous addition of N alone did not influence available N, instead reduced available N when compared to NPK treatments. INM is the best way of improving soil available nutrients and soil enzymatic activities for sustainable soil health.

Keywords- *Urease, Phosphatase, Dehydrogenase, Soil enzymes, Manures, Fertilisers*

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Introduction

The introduction of high yielding and high nutrient responsive crop varieties has increased the input requirement of nutrients. Although adoption of modern scientific techniques has increased the crop production significantly in the recent years, agricultural production has to be increased manifold and sustained at profitable levels to meet the requirement of ever increasing population [1]. Large quantum of fertilizers has to apply in soil by chemical form which may impact on soil health in log run. Interest in long-term field experiments as the suitable indicators of sustainability of agriculture has increased during last few decades the world over. These experiments indicate the extent to which yield and related parameters and the quality of ecosystem can be predicted. These are also capable of serving as an early warning system to detect problems that threaten future productivity [2]. Nearly 80 -90 percent of the soil processes are mediated by soil microbes in ecosystem [3]. Soil microbial population are the driving force behind regulating soil processes such as organic matter decomposition and nutrient cycling, it is imperative to have a better understanding of the factors that regulate its size, activity, and structure [4]. Soils containing a high microbial diversity are characteristic of a healthy soil-plant relationship, whereas those with low microbial diversity are characterized as an unhealthy soil that often hardly responds to environmental changes [5]. Microbial activities in soil, despite their importance in many of the soil processes, are frequently disturbed by altered soil enzyme activities as a result of agricultural exploitations and tillage practices. Disruption in soil microbial activity as shown by changes in levels of metabolic enzymes, can serve as an estimate of ecosystem disruption [6]. The different types of soil enzymes studied from various objectives of investigations, one oxidoreductase (dehydrogenase) and two hydrolases (phosphatase and urease) are thoroughly studied enzymes due to their specific importance in organic matter transformation processes, phosphorous cycle and agricultural practices.

Application of concentrate organic manure and inorganic fertilizer could significantly involve in urease and phosphatase enzyme activity in soil and it may lead to involved in N and P cycles [7]. Soil phosphatase hydrolyzes the ester bonds binding P to C (C-O-P) in organic matter and in this process inorganic P is released from organically bound P. Soils under phosphorus deficient both plants and microorganisms release phosphatase enzymes into the soil which have the potential to mobilize the P reserve [8]. Dehydrogenase enzyme oxidizes soil organic matter by transferring protons and electrons from substrates to acceptors. Soil dehydrogenase is considered as an indicator of overall microbial activity because it occurs intracellulary in all living microbial cells and is linked with microbial oxydoreduction processes [9]. However limited information is available on microbial diversity under long term experimentation of manure - fertilizer schedules under rice monoculture. With this background, the present study has undertaken to assess the effect of long term application of manures, fertilizers and integrated nutrient application practices on soil biological properties and their influence on availability of macro nutrients in rice crop.

Materials and methods

Site description and treatments

The permanent manurial experiment is in operation since 1975 at the Agricultural College and Research Institute, Madurai, Tamil Nadu (95°4' North latitude, 78°0' East longitude and 147 m above MSL) in sandy clay loam soil (Typic haplustalfs). The experiment was laid out in split plot design with two replications (Main plot : M_1 - No manure, M_2 - Farm yard manure (FYM) @ 12.5 t ha^{-1}, M_3 - Green leaf manure (GLM) @ 12.5 t ha^{-1}, M_4 - Urban compost (UC) @ 12.5 t ha^{-1}; Sub-plot - S_1 - Control (No, N, P and K); S_2 - N, S_3 - P, S_4 - K, S_5 - N + P, S_6 - N+ K, S_7 - P + K, S_8 - N + P + K. The organic manures *viz*., FYM, GLM and UC were applied and incorporated into the soil one week prior to planting, nitrogen, phosphorus and potassium is applied at the rates of 150, 50 and 50 kgha^{-1}, respectively in the form of urea, super phosphate and muriate of potash according to the treatments. The entire does of P was applied basally and N and K were applied in three equal splits *viz*., 1/3 as basal, 1/3 at active tillering stage and 1/3rd at panicle initiation stage of the corp. The present study was undertaken with the 57th rice crop raised in the experiment during *rabi* season 2014-15.

Methods employed for Analysis

Available N in soil was estimated by alkaline permanganate method [10]; available P by Colorimetry method [11], available K by Neutral Normal Ammonium Acetate method [12], and values were subjected to analysis of variance.

Soil samples

Preplanting and post harvest soil samples of the 57th crop were collected at 0-15 cm depth, air dried in shade, ground with wooden mallet, passed through 2 mm sieve and bagged in plastic bags and stored for the chemical analysis. Composite surface soil samples were collected during preplanting, active tillering, flowering and at post harvest stages at field moisture conditions in all the treatments. These moist samples were kept at 4ºC until the analysis were carried out, which took no longer than two days after sampling. These samples were analyzed for soil microbes *viz*., bacteria, fungi, actionmycets and enzymes *viz*., urease, phosphatase and dehydrogenase.

Initial characterization of the experimental soil

The soil of the experimental site was sandy clay loam in texture with pH of 7.24 and electrical conductivity of 0.30 dS m^{-1}(Table 1). The total N, P and K status of the soil were 0.08, 0.06 and 0.11 percent respectively. The available N, P and K status of the soil were 185, 25.5 and 254 kg ha^{-1} respectively. The cation exchange capacity (CEC) of the soil was 22.8 cmol (p+) kg^{-1}. The experimental soil belonged to the order Alfisol (Typic Haplustalf) as per the soil taxonomy.

Nutrient composition of the organic manures

Among the manures used during *rabi* 2014, the N content was the highest in GLM (2.72%), whereas it was 0.56 and 0.65 percent in FYM and UC, respectively (Table 2.). The UC registered the highest P content (0.56%) and the K content was the highest in GLM (1.53%).

Assay of soil enzymes

Urease

The urease activity in soils was measured according to the method prescribed by [13]. To 5 g of soil, 1ml of 0.2 ml urea solution was added as substrate and incubated at 37ºC for 2 hours. To the contents, 35 ml of KCl – Ag_2SO_4 solution was added and the ammoniacal-N released by steam distillation was determined by Bremner method [14]. The soil urease activity was expressed as g of ammoniacal-N released kg^{-1} soil hr^{-1}.

Phosphatase

The phosphatase activity in soil was estimated following the method of [13]. To 1g of soil sample, 3 ml of ρ-nitrophenol phosphate and 3.5 ml of 0.1M phosphate – malate buffer was added and incubated for 4 hours. After incubation, 1 ml of 0.5 M calcium and 4 ml of 0.5 M sodium hydroxide were added. The contents were filtered and the intensity of the colour developed was measured at 420 nm using a colorimeter. The phosphatase activity was calculated by using the standard graph prepared with ρ-nitrophenol and expressed as g of phenol released kg^{-1} soil hr^{-1}.

Dehydrogenase

The activity of dehydrogenase enzyme in soils was assayed according to the method prescribed by [15]. To 1g of soil sample, 10ml of phosphate buffer containing 1 percent 2, 3, 5-triphenyl tetrazolium chloride was added and incubated at room temperature for 15 hours. After incubation, 40 ml of 90 percent carbon tetrachloride was added and mixed well. The concentration of the triphenyl formazon (TPF) was estimated at 530 nm in a colorimeter. The activity of the dehydrogenase was calculated from the standard graph prepared with triphenyl teterzolium formazon and expressed as g of TPF released kg^{-1} soil hr^{-1}.

Statistical analysis

The data obtained from different experiments was analysed statistically to find out the effects due to various treatments and their interactions. Analysis of variance was calculated as suggested [16].

Results and Discussion

Long term effect of manure – fertilizer schedules on soil enzyme activities

Soil urease activity

Urease is an enzyme which is responsible for hydrolysis of urea in to CO_2 and NH_4+ in soil and also it is an important factor for survival of NH_4+ fertilizer oxidizers present in soil. The biogeochemical activities in terms of soil enzymes in their existence and distribution are affected by application of manures and fertilizer in a long run. Urease enzyme activity was significantly higher in application of organic manures in soil at tillering, flowering and post harvest stages of rice crop (Table 3). Among the manures, the maximum urease activity (173 µg of NH_4-N g^{-1} of dry soil h^{-1}) in soils was recorded in plots with GLM followed by FYM applied plots. Application of organic manure augmented the soil organic carbon reserve; its serves the nutrient input for added big quantity of organic and microbial carbon by natural manure. This might be that organic matters could increase microbial activity in the soil by provide better conditions for the accumulation of enzymes in the soil. The activity of urease, the enzyme that catalyzes the hydrolysis of urea and which is widely used in the evaluation of changes in soil quality for soil management increased with the application of compost [17]. Also, the enhanced level of soil enzyme activity due to addition of organic manures promotes the recycling of nutrients in the soil ecosystem [18]. For applying chemical fertilizers, the urease activity was ranged from 102 µg of NH_4-N g^{-1} of dry soil h^{-1} in unfertilized control to 142 µg of NH_4-N g^{-1} of dry soil h^{-1} in the treatment that received NPK at 150:50:50 kg ha^{-1}. Urease activity was decreased by application of chemical fertilizers alone because urea fertilizers and the reaction products have ammonium, causing microbial induction of urease activity. Application of chemical fertilizers (Conventional Tillage with 90 kg triple super phosphate ha^{-1} + 60 kg Urea ha^{-1}) significantly decreased urease activity, whereas adding organic manure (No Till with 2700 kg FYM ha^{-1} + 1250 kg compost ha^{-1}) resulted in an increase in its activity. While comparing treatments, the urease activity was significantly influenced by the combined application of manures and fertilizers, and the maximum being with the application of GLM along with recommended NPK [19]. Enzyme activities in the soil are closely related to organic matter content and application of balanced amount of nutrients and manures improve the MBC status of soils which corresponds to higher enzyme activity [20]. The activity of urease, the enzyme that catalyzes the hydrolysis of urea and which is widely used in the evaluation of changes in soil quality for soil management increased with the application of compost. The low urease activity found in the CF treatment (similar to that of the control treatment) was expected because the presence of inorganic forms of N makes the synthesis of the enzyme unnecessary. Higher urease activity probably resulted from an increase in soil organic matter content and microbial population, resulting in the secretion of urease, although no urea was applied.

Soil phosphatase activity

Phosphatase is an extracellular enzyme in soil as alkaline phosphatase which is responsible for converting orthophosphoric monoester to easy accessible orthophosphate ions in soil. This enzyme is produced by many organisms in the soil. It remove the phosphate molecule from organic compounds such as phospholipids and nucleic acids and the phosphate is cleaved it becomes soluble and can be taken up by the cell. Amendments of organic manures significantly influenced the phosphatase activity of soil at critical stages of rice crop (Table 4). Among the manures, the maximum phosphatase activity (134 mg PNP g^{-1} of dry soil h^{-1}) was recorded in plots that received GLM followed by FYM applied plots. This might be that application of organic amendments could increase the soil microbial carbon elevated the phosphatase activity in soil.

Soil phosphates could convert the insoluble orthophosphate molecules in to free ions that can be easily taken up by the plants. Soil amended with organic fertilizers (C and P), b-glucosidase was positively correlated with phosphatase activity (P< 0.001) and those enzyme activities with organic C (P <0.01), which explain that the inclusion of organic manures furnishes substrates for the both enzymes and improves microbial expansion [21]. By the application of chemical fertilizers the values was ranged from 106 Mg PNP g^{-1} of dry soil h^{-1} in unfertilized control to 132 Mg PNP g^{-1} of dry soil h^{-1} in the treatment that received NPK at 150:50:50 kg ha^{-1}. Comparing the treatments, chemical fertilizer application could reduce the phospatase activity in soil. The possible reason may be that chemical fertilizer inhibited the synthesis of phosphatase in soil. Application of manures could give the favorable environment for microbes using as substrate in soil. P fertilizer had a negative effect on alkaline phosphatase activity and dehydrogenase activity is highly sensitive to inhibiting effect associated with mineral fertilizer addition [22]. The phosphatase activity was significantly influenced by the combined application of manures and fertilizers, the maximum being with the application of GLM along with recommended NPK. Influence of compost addition and inorganic fertilizer treatment on soil biological and yield of crop under a cereal-legume on a Typic Haplaustalf increased alkaline phosphatase activities with addition of organic material whereas no significant influence of inorganic fertilizer treatment was observed [22,23].

Soil dehydrogenase activity

Soil dehydrogenases are the extracellular, Oxidoreductase enzyme which is responsible for the biological oxidation of soil organic matter (OM) by transferring hydrogen from organic substrates to inorganic acceptors. Soil amended with organic manures had significantly influenced the soil dehydrogenase activity at critical stages of rice crop compared to unmanured control (94 TPF Mg g^{-1} of dry soil h^{-1}) (Table 5). The dehydrogenase activity was ranged from 106 TPF Mgg^{-1} of dry soil h^{-1} to 132 TPF Mgg^{-1} of dry soil h^{-1} and GLM received higher DHA of 132 TPF Mgg^{-1} of dry soil h^{-1} in organic manure treatment. This due to that green leaf manure have higher nutrient content compared to other amendments and it could increase the soil microbial biomass and enzyme activities are closely related to OM content of the organic amendments. Also the metabolic ability of the microbial communities and their capacity to use substrates as based on the quality and type of the organic manures. This treatment was followed by GLM with P+K which was also on par with GLM with N+K application. The higher dehydrogenase activity due to addition of GLM could be attributed to increased microbial activity which is known to stimulate the dehydrogenase activity [25]. While comparing the stages, the higher dehydrogenase activity was observed in tillering stages in rice. Addition of FYM, GLM and Urban Compost over four decades increased the readily available C and N pools by the substrate induced microbial biomass in soil. Regarding the dehydrogenase activity was low in control (96 TPF Mg g^{-1} of dry soil h^{-1}) and maximum in NPK treated plots (128 TPF Mg g^{-1} of dry soil h^{-1}). A possible reason may be that, inorganic fertilizer application could inhibit the dehydrogense activity. The application of inorganic fertilizers had relatively less effect on soil enzymes activity than organic fertilizers [26, 27]. Also, noted that DHA usually reached higher level in the organic treatments [28].

Effect of manure – fertilizer schedules on soil available nutrient status

Soil amended with organic manures and nitrogenous fertilizers significantly influenced the available N status of the soil. Among the organic manures, application of GLM significantly recorded the maximum available N status (249 kg ha^{-1}) followed by that of FYM application (233 kg ha^{-1}) compared to unmanured control (184 kg ha^{-1}) (Table 6).This was due to GLM are rich in nutrient content, incorporation of GLM or other organic manures could get decomposed and nutrients are too available to the plants. Also amending GLM in soil reduced the bulk density and increased the OM and OM with soil N was positively correlated. Amending, organic manures had more significant effect on soil available N and the effect was more pronounced with the application of GLM @ 12.5 t ha^{-1} [29]. Among the inorganic fertilizers, application of nitrogenous fertilizers with or without P and K recorded higher available N status. The maximum availability of N (288 kg ha^{-1}) was recorded in the treatment that received NPK compared to unfertilized control (196 kg ha^{-1}). This may be attributed that application of chemical fertilizer did not improve the OM in soil; it could leads the less soil microbial biomass and reduces the nutrient catalytic capacity of the soil. Integration of organic manures and fertilizers significantly influenced the available N status, the maximum being with the application of GLM @ 12.5 t ha^{-1} and NPK @ 150:50:50kg ha^{-1}. This was affirmed that, integration of organic manures with chemical fertilizers could decrease the nutrient leaching and addition of organic manures induced more soil ammonium oxidizing bacteria or nitrifiers in soil. These nitrifiers could reduce the ammonium in to plant accessible form of NO_3- . Significant correlation of different forms of nitrogen was observed with continuous application of both inorganic fertilizers and green manuring [30].

Soil available P status

Olsen - P was significantly higher in treatments that received manures than in unmanured control (Table 7). The maximum P availability (42.8 kg ha^{-1}) was registered in the soils that received GLM followed by that of FYM application (33.4 kg ha^{-1}). This was affirmed that application of GLM could increase the soil phosphates enzyme lead to catalyses the organic P to more available orthophosphate free ions in soil solution. Also it builds up better rhizhopheric environment and secretion of organic acids had increased the native P more available to plants. The P desorption which could occur as a result of an interaction between organic molecule- complexes with FYM [31] on one side and pH changes on the other side [32]. However, inorganic treatments that received P with or without N and K recorded higher P availability and it was ranged from 23.2 in unfertilized control to 41.2 kg ha^{-1} in recommended NPK ha^{-1}. Integrated application of GLM (M_3) along with NPK followed by GLM with N+K recorded the maximum P available P status. A possible reason may be, organic manures are substrate for the soil macro fauna and the presence of organic acids in decomposed products of manure induced the P desorption from the fixed sites and clay – humus complexes. Evaluation of different green manures found that the utilization of phosphorous fertilizers had markedly enhanced it from 3% to 39% in treatments having green manure [33, 34]. Also, similar buildup of phosphorus was observed by [35] under a long term fertilizer experiment which received higher application of 150% NPK followed by the treatment that received 100% NPK + FYM.

Soil available K status

The available potassium content differed significantly due to various sources of organics in combination with inorganic fertilizers (Table 8). Like available nitrogen and phosphorus, the highest available K content (313 kg ha^{-1}) was recorded in the treatment that received GLM + 100% NPK at preplanting stage followed by FYM + 100% NPK (300kg ha^{-1}). The available K status of post harvest soils was considerably higher in these same treatments compared to that of unmanured and unfertilized control. The beneficial effects of GLM, FYM and UC on available K status may be ascribed to the direct potassium addition in the potassium pool of the soil. The higher available K was observed under conjoint use of organics and fertilizers than the sole use of fertilizer [36].

Conclusion

The present work proves that, amending organic manures and inorganic ferilisers may considerably alter the soil microbial communities by the way of changing soil physical and physico chemical properties such as soil fertility. Soil microbial and enzymatic properties were dependent mainly on SOC of the soil.

Table-1 *Basic properties of the experimental soil*

i) Mechanical analysis	Values	iii) Physico-chemical properties	Values
Texture class	Sandy clay loam	pH	7.24
		EC (dSm^{-1})	0.30
		CEC (c mol(p^+) kg^{-1})	22.8
ii) Physical properties		iv) Chemical properties	
Bulk density (Mg m^{-3})	1.28	Organic carbon (g kg^{-1})	6.4
Particle density (Mg m^{-3})	2.51	Alkaline $KMnO_4$ – N (kg ha^{-1})	185
Water holding capacity (%)	41.4	Olsen P (kg ha^{-1})	25.5
Volume expansion on wetting (%)	10.6	N N NH_4OAc-K (kg ha^{-1})	254

Table-2 *Composition of organic manures*

Organic manures	Nutrient content (%)								
	C	N	P	K	S	Zn ppm	Mn ppm	Cu ppm	Fe ppm
FYM	36.8	0.56	0.25	0.46	0.02	36.0	20.0	1.11	18
GLM	63.8	2.72	0.40	1.53	0.39	24.0	10.0	0.92	8.0
UC	55.4	0.65	0.56	0.40	0.42	26.0	13.0	1.40	16.0

Table-3 *Effect of long term manure and fertilizer application in urease activity (µg of NH_4-N g^{-1} of dry soil h^{-1})*

Treatments Sub/Main	Urease activity (µg of NH_4-N g^{-1} of dry soil h^{-1})															
	Tillering					Flowering					Post Harvest					Sub plot mean
	M_1	M_2	M_3	M_4	Mean	M_1	M_2	M_3	M_4	Mean	M_1	M_2	M_3	M_4	Mean	
S_1	97	137	181	126	135	82	122	166	111	120	42	62	96	51	51	102
S_2	128	159	211	154	163	113	144	196	139	148	73	84	126	79	79	130
S_3	115	142	196	146	149	100	127	181	131	135	60	67	111	71	71	118
S_4	122	153	203	151	157	107	138	188	136	142	67	78	118	76	76	125
S_5	134	164	218	156	168	119	149	203	141	153	79	89	133	81	81	134
S_6	136	168	221	159	171	121	153	206	144	156	81	93	136	84	84	137
S_7	108	148	188	138	146	93	133	173	123	130	53	73	103	63	63	113
S_8	140	176	228	162	177	125	161	213	147	161	85	101	143	87	87	142
Mean	123	156	206	149	158	107	141	191	134	143	68	81	121	74	74	
Main plot mean	M_1– 99				M_2 - 126					M_3 - 173					M_4– 119	

	Tillering		Flowering		Post harvest	
	SEd	CD(p=0.05)	SEd	CD(p=0.05)	SEd	CD(p=0.05)
M	2.38	5.68	2.64	5.34	0.72	1.43
S	2.45	5.97	2.48	5.87	0.37	1.78
M at S	4.19	8.26	4.39	8.63	1.64	3.24

Table-4 *Long term effect of manures and fertilizers on phosphatase activity of the soil*

Treatments Sub/Main	Phosphatase activity (µg of PNP g^{-1} of dry soil h^{-1})															Sub plot Mean
	Tillering					Flowering					Post Harvest					
	M_1	M_2	M_3	M_4	Mean	M_1	M_2	M_3	M_4	Mean	M_1	M_2	M_3	M_4	Mean	
S_1	123	145	156	140	141	93	115	126	110	111	52	73	82	65	68	106
S_2	151	162	171	157	160	121	132	141	127	130	78	89	93	82	86	125
S_3	142	158	167	153	155	112	128	137	123	125	71	87	90	78	82	120
S_4	144	161	169	156	158	114	131	139	126	128	73	90	90	81	84	123
S_5	148	153	178	148	156	118	123	148	118	126	77	82	95	73	81	121
S_6	154	164	175	159	164	124	134	145	129	134	73	91	92	84	86	128
S_7	140	156	165	151	153	110	126	135	121	123	69	85	87	76	79	118
S_8	160	168	184	163	169	130	138	154	133	139	79	92	98	86	89	132
Mean	145	158	171	157	157	115	128	141	123	127	72	86	91	78	82	
Main plot mean	M_1– 110			M_2– 124					M_3 – 134					M_4 - 119		

	Tillering		Flowering		Post harvest	
	SEd	CD(p=0.05)	SEd	CD(p=0.05)	SEd	CD(p=0.05)
M	2.11	6.71	2.32	4.22	1.09	2.14
S	1.63	3.35	2.65	4.68	1.45	2.65
M at S	3.72	8.98	3.58	7.98	2.46	4.69

Table-5 *Long term effect of manures and fertilizers on dehydrogenase activity of the soil*

Treatments Sub/Main	Dehydrogenase activity (TPF µg $g^{-1}$of dry soil h^{-1})															
	Tillering					Flowering					Post Harvest					
	M_1	M_2	M_3	M_4	Mean	M_1	M_2	M_3	M_4	Mean	M_1	M_2	M_3	M_4	Mean	Sub plot mean
S_1	90	122	138	120	118	65	99	116	98	95	51	81	98	75	76	96
S_2	112	118	146	133	127	88	96	124	107	104	82	80	106	91	90	107
S_3	122	138	156	136	138	98	115	134	111	115	90	92	109	95	97	116
S_4	108	127	144	115	124	84	105	122	92	101	78	88	103	82	88	104
S_5	118	143	165	136	141	92	120	142	113	117	88	95	107	94	98	118
S_6	106	114	146	124	123	82	92	123	102	100	75	85	105	79	86	103
S_7	115	148	174	127	141	91	122	151	106	118	87	102	114	74	92	117
S_8	132	153	182	140	152	108	128	154	117	127	94	107	120	98	105	128
Mean	113	133	156	128	133	89	110	133	106	109	81	91	108	86	91	
Main plot mean	M_1 – 94				M_2 - 111					M_3 – 132					M_4– 106	

	Tillering		Flowering		Post harvest	
	SEd	CD(p=0.05)	SEd	CD(p=0.05)	SEd	CD(p=0.05)
M	1.31	4.16	1.67	2.82	1.43	2.38
S	1.35	2.78	1.83	3.14	1.64	2.87
M at S	2.85	6.53	3.27	6.34	2.81	5.12

Table-6 Effect *of manure - fertilizer schedules on available N status (kg ha^{-1}) of the soil*

Treatments Sub/Main	Pre planting stage					Post Harvest stage					
	M_1	M_2	M_3	M_4	Mean	M_1	M_2	M_3	M_4	Mean	Sub plot mean
S_1	149	206	219	190	191	159	216	229	200	201	196
S_2	185	223	246	220	219	195	233	256	230	229	224
S_3	153	209	223	195	195	163	219	233	205	205	200
S_4	158	213	228	210	202	168	223	238	220	212	207
S_5	186	235	248	225	224	196	245	258	235	234	229
S_6	192	236	256	230	229	202	241	266	240	237	233
S_7	150	210	217	178	189	160	215	227	188	198	194
S_8	255	290	310	276	283	265	301	321	286	293	288
Mean	179	228	243	216	216	189	237	254	226	226	
Main plot mean	M_1 – 184			M_2 - 233			M_3 - 249			M_4 - 221	

	Pre planting stage		Post Harvest stage	
	SEd	CD(P=0.05)	SEd	CD(P=0.05)
M	5.1	8.4	4.3	9.1
S	5.9	8.6	4.8	9.4
M at S	7.5	12.3	6.3	12.8

Table-7 *Effect of manure - fertilizer schedules on available P status (kg ha^{-1}) of the soil*

Treatments Sub/Main	Pre planting stage					Post Harvest stage					Sub plot mean
	M_1	M_2	M_3	M_4	Mean	M_1	M_2	M_3	M_4	Mean	
S_1	10.1	16.8	23.9	16.6	16.8	22.1	29.8	36.9	29.6	29.6	23.3
S_2	18.1	20.1	33.5	20.8	23.1	31.3	33.1	46.5	33.8	36.1	30.0
S_3	23.1	32.1	37.1	26.1	29.6	36.3	45.6	50.1	39.1	42.7	36.2
S_4	17.3	24.4	32.6	18.7	23.2	30.3	37.4	45.6	31.7	36.2	29.7
S_5	21.3	33.6	39.1	27.9	30.4	34.1	46.5	52.1	40.9	43.4	37.0
S_6	16.8	19.1	34.4	21.8	23.0	29.8	32.1	48.4	34.8	36.2	30.0
S_7	19.4	34.0	40.1	29.0	30.6	32.4	47.0	53.1	42.0	43.6	37.1
S_8	24.3	35.0	46.1	30.3	33.9	37.3	47.9	65.1	43.3	48.4	41.1
Mean	18.8	26.8	35.8	23.9	26.3	31.7	39.9	49.7	36.9	39.5	
Main plot mean	M_1- 25.3			M_2- 33.4			M_3- 42.8			M_4- 30.4	

	Pre planting stage		Post Harvest stage	
	SEd	CD (P=0.05)	SEd	CD (P=0.05)
M	0.62	1.21	0.81	1.70
S	0.65	1.31	0.92	1.82
M at S	1.32	2.73	1.82	3.78

Table-8 *Effect of manure - fertilizer schedules on available K status (kg ha^{-1}) of the soil*

Treatments Sub/Main	Pre planting stage					Post harvest stage					Sub plot mean
	M_1	M_2	M_3	M_4	Mean	M_1	M_2	M_3	M_4	Mean	
S_1	190	205	230	200	206	205	220	245	213	221	214
S_2	205	207	295	212	230	220	222	210	227	220	225
S_3	214	217	244	216	223	229	232	259	231	238	231
S_4	261	223	275	269	257	274	281	290	284	282	270
S_5	213	285	234	205	234	228	238	249	220	234	234
S_6	265	285	283	278	278	280	300	298	293	293	286
S_7	240	296	295	280	278	265	311	310	295	295	287
S_8	280	300	313	293	299	295	315	328	308	312	306
Mean	234	252	271	245	250	250	265	274	259	262	
Main plot mean	M_1- 242			M_2- 259			M_3- 273			M_4- 252	

	Pre planting stage		Post Harvest stage	
	SEd	CD(P0.05)	SEd	C.D(P=0.05)
M	5.5	11.4	6.3	12.2
S	6.2	12.6	6.7	13.8
M at S	9.3	18.3	8.3	19.1

Impacts were more pronounced in amending organic manures especially GLM. Integrated application of GLM @ 12.5 t ha^{-1} along with N fertilizer with or without P and K registered the maximum urease (20.61 %), phosphatase (15.21%) and DHA (24.17%) activity over control. The reason for this hypothesis is that organic manures could improve the soil structure and increases the soil available nutrient by the way of microbes catalyzes the organic ions in to plant accessible free ions in soil solution. Application of inorganic fertilizers alone could reduce enzymes activity especially dehydrogenase which is important oxidoreductase enzyme responsible for breakdown of OM and nutrient cycling in soil. It is concluded that, integration of chemical fertilizers NPK with organic manures especially GLM could improve the soil biochemical properties and soil available nutrients status and small buildup after harvest of the rice crop in a long run.

Application of Research: This study has reflected on the sustainable agriculture towards soil health

Research Category: Agricultural Sciences

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***Research Guide or Chairperson of research: Dr B. Bakiyathu Saliha**
University: Tamil Nadu Agricultural University, Coimbatore, 641 003, Tamil Nadu
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Study area / Sample Collection: Agricultural College and Research Institute, Madurai, Tamil Nadu

Cultivar / Variety name: Rice - *Oryza sativa*

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