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Effect of integrated nutrient management practices on soil enzyme activity and microbial population in paddy

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Abstract

A field experiment was conducted at College Farm, S.V. Agricultural College, Tirupati during kharif, 2022-23 to study the effect of integrated use of organic manures, green manures, biofertilizers and inorganic fertilizers on enzymatic activity and microbial population in paddy. The experiment was laid out in a split-plot design with seven main plots and three sub plots with three replications. The main plots comprised of seven different combination of organic manures viz., M1: FYM @ 10 t ha-1 M2: Green manure seed (dhaincha) @ 25 kg ha⁻¹), M₃: FYM @ 5 t ha⁻¹ + Green manures seed @ 12.5 kg ha⁻¹, M₄:FYM @ 10 t ha⁻¹ + Azospirillum + PSB + KRB @ 1.25 l ha⁻¹ each, M₅: Green manure seed @ 25 kg ha⁻¹ + Azospirillum + PSB + KRB @ 1.25 l ha⁻¹ each, M₆: FYM @ 5 t ha⁻¹ + Green manures @ 12.5 kg ha⁻¹ + Azospirillum + + KRB @ 1.25 l ha-1 each and M7:Control. Sub-plots comprised of three levels of fertilizers viz., S1: 50% RDF, S2:75% RDF and S3:100% RDF. The enzyme activity was estimated and total microbial population was recorded in soil at panicle initiation and harvest stage of paddy crop. Among the various organic combinations, significantly the highest urease, dehydrogenase and phosphatase activity was recorded with FYM @ 5 t ha⁻¹ + green manures @ 12.5 kg ha⁻¹ + Azospirillum + KRB @ 1.25 l ha⁻¹ each which was on par with the application of green manures @ 12.5 kg ha⁻¹ + Azospirillum + PSB + KRB @ 1.25 l ha⁻¹ each (M_5) and FYM @ 10 t ha⁻¹ + Azospirillum + PSB + KRB @ 1.25 l ha⁻¹ each (M_4) and the lowest was recorded with control (M_7) . However, among the fertilizer levels, the highest enzyme activity was obtained with 100% RDF (S₃) which was comparable with 75% RDF (S₃) and 50% RDF (S₃). Similarly, among various organic combinations, significantly the highest total bacteria, fungi and actinomycetes population were recorded with FYM @ 5 t ha⁻¹ + green manures @ 12.5 kg ha⁻¹ + Azospirillum + KRB @ 1.25 l ha⁻¹ each (M₆) which was on par with the application of green manures @ 12.5 kg ha⁻¹ + Azospirillum + PSB + KRB @ 1.25 l ha⁻¹ each (M₅) followed by FYM @ 10 t ha⁻¹ + Azospirillum + KRB @ 1.25 l ha⁻¹ each (M₄) and the lowest were recorded with control (M7). However, among the fertilizer levels, the highest microbial population was recorded with 100% RDF (S₃) which was comparable with 75% RDF (S₂) and 50% RDF (S1). The enzyme activity and microbial population were highest in combined application of FYM @ 5 t ha⁻ + green manures @ 12.5 kg ha⁻¹ + Azospirillum + KRB @ 1.25 l ha⁻¹ each along with application of 75% RDF (M6S2) at both PI and harvest of paddy.

Keywords: INM practices, soil enzyme activity, total microbial population, paddy crop

Introduction

Rice (*Oryza sativa* L.) is the principal food crop for the world's billions of people. It plays a vital role in our national food security. In India, it is grown in an area of 46.28 million hactares with a production of 129.47 million tonnes and productivity of 2798 kg ha⁻¹. In Andhra Pradesh, it is grown in an area of 2.23 million hactares with a production of 7.76 million tonnes and productivity of 3392 kg ha⁻¹ (www.indiasta.com., 2022-23).

Rice based cropping systems are the major production systems contributing to food production. Current crop production systems are characterized by inadequate and imbalanced uses of fertilizers. Crop demand for nutrients is met by a combination of inherent soil fertility and externally applied nutrients. For high yielding crops with high rates of dry matter accumulation removing higher rates of nutrients such as rice and maize, soil must allow unrestricted root growth be able to absorb nutrients at the rate for maximum growth. In order to cope with the food demand of a growing population, 60 percent increase in rice production will be necessary

during the next 25 years (Arth and Frenzel, 2000)^[1]. To achieve this goal, NPK fertilization has to be done at nearly 3 fold from the present level. But escalating prices of industrial fertilizers and their possible degradation effect on soil health and pollution to environment warrants the need for excess use of chemical fertilizers. In this context, adoption of integrated nutrient management involving organic and inorganic sources is the best nutrient technology available.

Usage of imbalanced fertilizers badly influences production potential and soil health. Integrated nutrient management will not only sustains the crop production but also be effective in improving soil health and enhancing nutrient use efficiency. Enzyme activities are considered as an index of microbiological activity. A better understanding of the role of these soil enzymes in the ecosystem could provide a unique opportunity for an integrated biological assessment of soils due to their crucial role in several soil biological activities, their ease of measurement, and their rapid response to the changes in soil management. Enzyme levels in soil systems vary in amounts primarily due to the fact that each soil type has different amounts of organic matter, composition and activity of living organisms and intensity of the biological processes. Since rice grows in the interactive ecosystem involving soil - microorganism - rice and atmosphere, rice development consequentially affect soil microorganisms and soil enzymatic activities (Bhavani et al., 2017)^[2]. Complementary use of organic and biological sources of plant nutrients along with chemical fertilizer is of great importance for the maintenance of soil health and productivity, especially under intensive cropping system. There is immense need to exploit the alternate source of nutrients viz., organic manure, green manures and biofertilizers, to sustain the productivity, soil health and soil fertility with more environment friendly nutrient management system. It provides balanced nutrition to crops and minimizes the antagonistic effects resulting from hidden deficiencies and nutrient imbalance. Improves and sustains the physical, chemical and biological functioning of soil.

Soil microorganisms play a key role in the maintenance of soil function or soil quality because they contribute to soil structure development and decomposition of organic manures. The boosted microbial activity increases the rate of readily available forms of nutrients for plant and synergistic effect on plant growth through improved enzyme activity. These changes have significant effect on quality and productive capacity of soil. Keeping in view the importance of integrated application of nutrients through various sources, the present study was taken up to know the changes in soil enzyme activity and total microbial population in paddy.

Materials and Methods

The experiment was conducted during *kharif*, 2022-23 at S.V. Agricultural College, Tirupati, Acharya N.G. Ranga Agricultural University, which is geographically situated at 13° 36' 761" N latitude and 79° 20' 704" E longitude with an altitude of 182.9 m above the mean sea level, which falls under Southern Agro Climatic Zone of Andhra Pradesh. The soil of experimental site was sandy clay loam in texture, neutral in reaction, low in electrical conductivity, low in organic carbon low in available nitrogen, medium in available phosphorus and available potassium. The experiment was laid out in a split-plot design with seven main plots and three subplots replicated three times. The main plots comprises seven combinations of various organic sources *viz.*, M₁: FYM @ 10 t ha⁻¹ (0.70, 0.25 and 0.57% N, P

and K applied two weeks before transplanting), M₂: Green manure seed (dhaincha) @ 25 kg ha⁻¹, M₃: FYM @ 5 t ha⁻¹ + Green manures seed @ 12.5 kg ha⁻¹ (3.40, 0.62 and 1.40% N, P and K was incorporated at flowering stage before transplanting of rice), M₄: FYM @ 10 t ha⁻¹ + *Azospirillum* + PSB + KRB @ 1.25 1 ha⁻¹ each, M₅: Green manure seed @ 25 kg ha⁻¹ + *Azospirillum* + PSB + KRB @ 1.25 1 ha⁻¹ each, M₆: FYM @ 5 t ha⁻¹ + Green manures @ 12.5 kg ha⁻¹ + *Azospirillum* + PSB + KRB @ 1.25 1 ha⁻¹ each, M₆: FYM @ 5 t ha⁻¹ + Green manures @ 12.5 kg ha⁻¹ + *Azospirillum* + PSB + KRB @ 1.25 1 ha⁻¹ each, M₇: Control. Whereas, the subplots comprises three vevele of inorganic fertilizers *viz.*, S₁: 50% RDF, S₂: 75% RDF and S₃: 100% RDF.

The RDF for rice crop in Southern Agro-Climatic Zone of Andhra Pradeshi is 120-60-40 kg ha-1. Nitrogen was applied in three equal splits in the form of urea for kharif rice (transplanting, tillering and panicle initiation), while phosphorus was applied entirely as basal through SSP and potassium in two equal splits (as basal and at panicle initiation stage) was applied though MOP. The area selected for raising nursery was thoroughly ploughed and leveled. Well-filled seed of rice cultivars NLR-34449 seed was broadcasted uniformly over puddled area @ 5 kg per 40 m2 area and the nursery was raised under irrigation up to the age of 25 days. The experimental field was ploughed twice by tractor drawn cultivator followed by a rotavator to obtain required tilth. The seedlings of 25 days were transplanted in all the plots at 2-3 seedlings hill⁻¹ with a spacing of 20 cm x 15 cm. The enzyme activity and total microbial population was recorded at both PI and harvest of paddy crop by following standard procedures as mentioned here. Urease activity was estimated by quantifying the rate of release of NH4⁺ - N from the hydrolysis of urea as described by Bremner and Douglas (1971)^[3]. Five grams of soil sample was taken in to 50 ml capacity glass tube and to that which 5 ml of urea was added. Cap the bottle and incubate at 37 °C. After 5 hours, remove the cap and add 50 ml of 2M KCI-PMA solution. Cap the bottle again, and shake it for 1 h. Filter the soil suspension under suction through Whatman no. 42 filter paper. Pipette an aliquot (1-2 ml) of the extract containing up to 200 mg l⁻¹ of urea into a 50 ml volumetric flask. Make the volume to 10 ml with 2M KCI-PMA solution, and add 30 ml of the colouring reagent. Swirl the flask for few seconds and place it in a bath of boiling water. After keeping 30 minutes on the water bath, remove the flasks and cool immediately in cold water (using ice) for 15 min. Make the volume to 50 ml with water and mix thoroughly. Measure the absorbance of red colour developed at 527 nm using spectrophotometer. The urease activity was expressed in terms of μg of NH_4^+ - N released $g^{-1} h^{-1}$.

Dehydrogenase activity in the soil sample was determined by following the procedure as described by Casida et al. (1964)^[4]. Exactly 5 grams of soil was taken in a screw cap test tube to which one ml of 3 percent aqueous solution of 2,3,5- triphenyl tetrazolium chloride (TTC), one ml of one percent glucose solution and 2.5 ml of distilled water (sufficient to leave a thin film of water above the soil layer) were added. The test tubes were stoppered with rubber bands and incubated at 30°C for 24 hours. At the end of incubation, the contents of the tube were rinsed down into a small beaker and converted into slurry by adding 10 ml methanol, the slurry was filtered through Whatman No. 1 filter paper. Repeated rinsing of soil with methanol was continued till filtrate was free of red colour. The intensity of red colour was measured at 485 nm wave length, against a methanol blank in a spectrophotometer. The concentration of formazon formed in the soil sample was determined using graded concentrations of formazon. The results were expressed in

milligram of triphenyl formazon (TPF) formed per gram of soil per day mg of TPF g⁻¹ day⁻¹. The procedure of Tabatabai and Bremner (1969) ^[23] and Evazi and Tabatabai (1977) ^[5] were adopted for the assay of acid and alkaline phosphatase activities. Soil samples (1.0 g) were taken in 100 ml Erlenmeyer flasks to which 0.2 ml toluene, 4.0 ml of modified universal buffer (pH 6.5 for assay of acid phosphatase and pH 11.0 for alkaline phosphatase) and 1.0 ml of *p*-nitrophenyl phosphate solution made in the same buffer were added. The Erlenmeyer flasks were swirled for a few seconds to mix the contents, stoppered and placed in an incubator at 37 °C for an hour. The stoppers were removed and 1.0 ml of 0.5 M CaCl₂ and 4 ml of 0.5 M NaOH were added to deactivate the enzyme and extract. The pnitrophenol liberated. The Erlenmeyer flasks were swirled for few seconds and the soil suspension was filtered through Whatman No.2v folded filter paper. The absorbance of the vellow colour of *p*-nitrophenol liberated due to the hydrolysis of the substract (p-nitrophenol phosphate) by the enzymes was measured at 420 nm by using a spectrophotometer against reagent blank. Controls were run simultaneously following the same procedure except adding 1.0 ml p-nitrophenol phosphate after addition of 1.0 ml of 0.5 M CaCl₂ and 4.0 ml of 0.5 M NaOH. Phosphatase activity was expressed as ug of pnitrophenol released g⁻¹ soil h⁻¹.

Microbial mass in the soil was normally expressed in colony forming units per gram of soil (CFU g⁻¹ of soil). The viable count for bacteria, fungi and actinomycetes was done on Nutrient Agar (NA), Martin's Rose Bengal Agar (RBA) and Ken-knight and Munaier's (KM) mediums, respectively. One gram of soil sample was taken, then added to 9 ml of sterile distilled water and shaken well for 10-15 minutes to obtain uniform suspension. This gives dilution of 1:100 (10⁻²). One ml of supernatant was transferred from 10⁻² dilution to 9 ml blank with sterile 1.0 ml pipette, which gave a dilution of 10^{-3} . The above process was repeated till the desired dilutions were obtained. The respective medium was melted at 42° and about 20 ml of medium was poured in each petri-plate and allow to solidify the soil suspension was spread on the solidified medium by taking 1ml of soil water suspension with 1ml micro pipette it (1ml) and was further placed into incubator after spreading. All these steps were carried out in the Laminar air flow chamber to avoid contamination. After incubation, colonies were developed and the viable count of soil microbes were counted (Tate, 1995) [24]

Results and Discussion

Effect of INM on soil enzyme activity

Data on urease, dehydrogenase, acid and alkaline phosphatase acitivity in soil at panicle initiation and harvest stages of paddy was presented in tables 1, 2 and 3. Enzyme activity was decreased with advancement in age of crop from panicle initiation to harvest.

Urease activity

Urease activity was significantly influenced by INM practices at both the stages of paddy crop. Among the manures, at both stages, significantly the highest urease activity (78.12 and 71.06 μ g NH₄ + g⁻¹ 2 hrs⁻¹ at PI and harvest, respectively) was recorded with the application of FYM @ 5 t ha⁻¹ + green manures @ 12.5 kg ha⁻¹ + *Azospirillum* + KRB @ 1.25 l ha⁻¹ each (M₆) which was on par with the application of green manures @ 12.5 kg ha⁻¹ + *Azospirillum* + PSB + KRB @ 1.25 l ha⁻¹ each (M₅) and FYM @ 10 t ha⁻¹ + *Azospirillum* + PSB + KRB @ 1.25 l ha⁻¹ each

(M₄) and the lowest (39.44 and 28.71 µg NH₄ ⁺ g⁻¹ 2 hrs⁻¹ at PI and harvest, respectively) was recorded with control (M₇). However, among the fertilizer levels, significantly the highest urease activity (65.96 and 57.31 06 µg NH₄ + g⁻¹ 2 hrs⁻¹ at PI and harvest, respectively) was obtained with 100% RDF (S₃) which was comparable with 75% RDF (S₂) and 50% RDF (S₁).

The interaction between application of various organic sources and inorganic fertilizers on urease activity at both stages of paddy crop was found significant. The highest uresae activity was recorded in combined application of FYM @ 5 t ha⁻¹ + green manures @ 12.5 kg ha⁻¹ + *Azospirillum* + KRB @ 1.25 l ha⁻¹ each along with application of 75% RDF (M6S2) at both PI and harvest of paddy.

Higher urease activity in the treatments that received organics might be due to promotion of biological and microbial activities which accelerated the breakdown of organic substances in the added manures. The higher organic matter levels in the farmyard manure and green manuring treatments might have provided a more favorable environment for the accumulation of enzymes in soil matrix, since soil organic constituents were thought to be important in forming stables complexes with free enzymes (Ramalakshmi *et al.*, 2011 and Vandana *et al.*, 2012) ^[20, 25]. These results were in consonance with Yadav *et al.* (2013) ^[26] and Reddy *et al.* (2016) ^[21].

Dehydrogenase activity

Dehydrogenase activity was significantly by INM practices at both the stages of paddy crop. Among manures, at both stages, significantly the highest dehydrogenase activity (99.89 and 87.44 µg TPF g⁻¹ soil day⁻¹ at PI and harvest, respectively) was recorded with application of FYM @ 5 t ha⁻¹ + green manures @ 12.5 kg ha⁻¹ + Azospirillum + KRB @ 1.25 l ha⁻¹ each (M₆) which was on par with the application of green manures @ 12.5 kg ha⁻¹ + Azospirillum + PSB + KRB @ 1.25 l ha⁻¹ each (M₅) and FYM @ 10 t ha⁻¹ + Azospirillum + PSB + KRB @ 1.25 l ha⁻¹ each (M₄) and the lowest (65.78 and 48.33 μ g TPF g⁻¹ soil day ⁻¹ at PI and harvest, respectively) was recorded with control (M_7) . However, among the fertilizer levels, significantly the highest dehydrogenase activity (90.31 and 75.55 µg TPF g⁻¹ soil day⁻¹ at PI and harvest, respectively) was obtained with 100% RDF (S₃) which was comparable with 75% RDF (S_2) and 50% RDF (S_1). The interaction between application of various organic sources

and inorganic fertilizers on dehydrogenase activity at both stages of paddy crop was found significant. The highest dehydrogenase activity was recorded in combined application of FYM @ 5 t ha⁻¹ + green manures @ 12.5 kg ha⁻¹ + *Azospirillum* + KRB @ 1.25 1 ha⁻¹ each along with application of 75% RDF (M6S2) at both PI and harvest of paddy.

The results of the present study revealed that application of balanced nutrients by combined use of organics and inorganics improved the organic matter status of soil which in turn enhanced dehydrogenase activity (Masto et al., 2006)^[13]. This might be due to the addition of organic sources which might have created environment conducive for formation of humic acid, stimulating the activity of soil microorganisms resulting in an increase in DHA of the soil. These results were coincided with the findings of Bajpai et al. (2006) and Prasad et al. (2010) ^[18]. Latha et al. (2019)^[10] also reported that application of FYM along with biofertilizers significant in improving dehydrogenase activity as it stimulated microbial population. Being chief carbon source, the organic sources supplemented through INM provided energy for soil microorganisms, and increased number of pores, which were considered important in soil-water-plant

relationships and maintained good soil structure accompanied by better dehydrogense activity. Similar results were also reported by Rai and Yadav (2011)^[19]. Lee et al. (2004)^[11] also reported that soil treated with organic manure showed higher level of dehydrogenase activity as compared to mineral fertilizers applied soil. Build-up of dehydrogenase activity in organics and inorganics treated plots may be due to increased organic carbon content and other nutrient sources that have contributed to the increased soil microorganisms which increased the dehydrogenase activity. These results were coincided with the findings of Mali et al. (2015)^[12] and Mounika et al. (2021)^[16].

Phosphatase activity

Acid and alkaline phosphatase activity was significantly by organic sources and fertilizer levels. However their interaction effect was non-significant during both the stages of paddy. Among manures, at both stages, the highest phosphatase activity (88.78, 76 and 118.21, 103.50 µg *p*-nitrophenol g⁻¹ h⁻¹ at PI and harvest, respectively) was recorded with application of FYM @ 5 t ha⁻¹ + green manures @ 12.5 kg ha⁻¹ + Azospirillum + KRB @ 1.25 l ha⁻¹ each (M_6) which was on par with the application of and green manures @ 12.5 kg ha⁻¹ + Azospirillum + PSB + KRB @ 1.25 l ha⁻¹ each (M₅) and FYM @ 10 t ha⁻¹ + Azospirillum + PSB + KRB @ 1.25 l ha⁻¹ each (M₄) and the lowest (53.33, 36.02 and 75.70, 58.96 μ g *p*-nitrophenol g⁻¹ h⁻¹ at PI and harvest, respectively) was recorded with control (M7) However, Among fertilizers levels, the highest acid and alkaline phosphatase activity (79.24, 64.79 and 106.64, 79.57 µg *p*-nitrophenol g⁻¹ h⁻¹ at PI and harvest, respectively) was obtained with 100% RDF (S₃) which was comparable with 75% RDF (S₂) and 50% RDF (S₁).

The higher acid and alkaline phosphatase activity in soil under organic nutrient management practices might be due to increased decomposition process and thereby increased the microbial activity. Addition of of organic amendments which would have favoured more microbial populations and this ultimately reflected on more enzymatic activity. Sriramachandrasekharan and Ravichandran (2011)^[22]. Addition of inorganic ferilizers along with biofertilizers increasing population of micro organisms due to increased availability of substrates which helps to release of enzymes of extra cellular origin (Kejiya et al., 2023)^[8].

Microbial population (bacteria, fungi and actinomycetes)

Data on microbial population in soil at panicle initiation and harvest stages of paddy of was presented in the Table 4. Microbial population had decreased from panicle initiation to harvest. Microbial population was significantly influenced by organic sources and fertilizer levels. However their interaction effect was non-significant during both the stages of paddy.

Total bacteria

Among the manures, at both the stages, significantly the highest bacteria population (63.46 and 48.33×10^6 CFU g⁻¹ soil at PI and harvest, respectively) was recorded with FYM @ 5 t ha⁻¹ + green manures @ 12.5 kg ha⁻¹ + *Azospirillum* + KRB @ 1.25 l ha⁻¹ each (M₆) which was on par with the application of green manures @ 12.5 kg ha⁻¹ + *Azospirillum* + PSB + KRB @ 1.25 l ha⁻¹ each (M₅) followed by FYM @ 10 t ha⁻¹ + *Azospirillum* +

KRB @ 1.25 l ha⁻¹ each (M₄) and the lowest was recorded with control (32.23 and 22.89 $\times 10^6$ CFU g⁻¹ soil at PI and harvest, respectively). However, among the fertilizer levels, significantly the highest bacterial population (54.47 and 41.49 $\times 10^6$ CFU g⁻¹ soil at PI and harvest, respectively) was obtained with 100% RDF (S₃) which was comparable with when compared to 75% RDF (S₂) and 50% RDF (S₁).

Total fungi

Among the manures, at both the stages, significantly the highest fungi population (24.22 and 18.56 ×10³ CFU g⁻¹ soil at PI and harvest, respectively) was recorded with FYM @ 5 t ha⁻¹ + green manures @ 12.5 kg ha⁻¹ + *Azospirillum* + KRB @ 1.25 l ha⁻¹ each (M₆) which was on par with the application of green manures @ 12.5 kg ha⁻¹ + *Azospirillum* + PSB + KRB @ 1.25 l ha⁻¹ each (M₅) followed by FYM @ 10 t ha⁻¹ + *Azospirillum* + KRB @ 1.25 l ha⁻¹ each (M₅) followed by FYM @ 10 t ha⁻¹ + *Azospirillum* + KRB @ 1.25 l ha⁻¹ each (M₄) and the lowest were recorded with control (13.78 and 9.67 89 ×10³ CFU g⁻¹ soil at PI and harvest, respectively). However, among the fertilizer levels, significantly the highest fungi population (20.86 and 16.18×10³ CFU g⁻¹ soil, 24.59 at PI and harvest, respectively) was obtained with 100% RDF (S₃) which was comparable with 75% RDF (S₂) and 50% RDF (S₁).

Total actinomycetes

Among the manures, at both the stages, significantly the highest actinomycetes population (28.44 and 21.64×10^4 CFU g⁻¹ soil at PI and harvest, respectively) was recorded with FYM @ 5 t ha⁻¹ + green manures @ 12.5 kg ha⁻¹ + *Azospirillum* + KRB @ 1.25 l ha⁻¹ each (M₆) which was on par with the application of green manures @ 12.5 kg ha⁻¹ + *Azospirillum* + PSB + KRB @ 1.25 l ha⁻¹ each (M₅) followed by FYM @ 10 t ha⁻¹ + *Azospirillum* + KRB @ 1.25 l ha⁻¹ each (M₄) and the lowest (16.00 and 10.28 × 10⁴ CFU g⁻¹ soil at PI and harvest, respectively) was recorded with control (M₇) was obtained with 100% RDF (S₃) which was comparable with 75% RDF (S₂) and 50% RDF (S₁).

Among the fertilizer levels, the treatment S₃- 100% RDF (54.47 and 41.49×10⁶ CFU g⁻¹ soil, 20.86 and 16.18×10³ CFU g⁻¹ soil, 24.59 and 18.66 $\times 10^4$ CFU g⁻¹ soil) recorded significantly higher bacteria, fungi and actinomycetes population when compared to 75% RDF (S₂) and 50% RDF (S₁) at flowering and harvest stage Microbial population had decreased from panicle initiation to harvest. These results coincided with Meghadubey et al. (2015) ^[14]. Islam and Borthakur (2016) ^[6] also found that the microbial population was the highest at panicle initiation and lowest at harvest. This might be attributed to increase in the root exudates in the panicle initiation in high moisture status leading to more intense microbial activity which gradually decline when the crop attained maturity to harvest stage. Organic nutrient management contributed significantly in enhancing the microbial count might be due to synergistic effect of biofertilizers with added organic manurial components having low C/N ratio, which in turn, induced high degree of mineralization than immobilization which consequently provided sufficient nutrients for the proliferation of microbes and their activities in terms of enzymes (Kumari et al., 2017)^[9]. Similar reported by Rai and Yadav, (2011)^[19], Monikarana et al. (2015)^[15], Nitin et al. (2015)^[17] and Jadhav et al. (2016)^[7].

Table 1: Effect of INM practices on soil urease activity (µg NH4 + g-1 2 hrs-1) at panicle Initiation and harvest stage of paddy

Treatments	Panicle initiation				Harvest								
Main plots	Fertilizer levels			Fertilizer levels									
	S1	S ₂	S 3	Mean	S 1	S ₂	S 3	Mean					
M ₁ : FYM @ 10 t ha ⁻¹	49.72	54.47	57.84	54.01	40.16	44.80	48.17	44.38					
M_2 : Green manure seed @ 25 kg ha ⁻¹	53.62	59.29	60.96	57.96	43.95	49.62	51.30	48.29					
M ₃ : FYM @ 5 t ha ⁻¹ + Green manures seed @ 12.5 kg ha ⁻¹	58.87	61.64	63.29	61.27	50.21	52.97	54.29	52.49					
$M_{4:}$ FYM @ 10 t ha ⁻¹ + Azospirillum + PSB + KRB @ 1.25 l ha ⁻¹ each	70.69	71.90	72.68	71.76	64.02	64.90	65.68	64.87					
M ₅ : Green manure seed @ 25 kg ha ⁻¹ + Azospirillum + PSB + KRB @ 1.25 l ha ⁻¹ each	71.07	75.93	77.50	74.83	64.07	68.93	70.50	67.83					
M ₆ : FYM @ 5 t ha ⁻¹ + Green Manures @ 12.5 kg ha ⁻¹ Azospirillum + PSB + KRB @	74 51	70.07	80.78	78 12	67 51	72.07	73 58	71.06					
1.25 l ha ⁻¹ each	74.31	4.51 79.07	1 79.07	19.07	19.07	19.07	79.07	80.78	76.12	07.51	72.07	75.58	/1.00
M ₇ : Control	27.38	42.30	48.65	39.44	16.52	31.97	37.65	28.71					
Mean	57.98	63.51	65.96		49.49	55.04	57.31						
	SEm +		CD (P=0.05)		SEm +		CD (P=0.05)						
М	2.30		7.10		2.23		6.96						
S	0.77		2.22		0.76		2.21						
S at M	2.03		6.26		2.02		6.21						
M at S	3.46		10.45		3.40		10.26						

Table 2: Effect of integrated nutrient management practices on dehydrogenase (µg TPF g⁻¹ soil day⁻¹) activity at panicle initiation and harvest stage of paddy

Treatments	Panicle initiation				Harvest				
Moin plata		Fertilizer levels				Fertilizer levels			
S1	S ₂	S 3	Mean	S 1	S ₂	S 3	Mean		
M ₁ : FYM @ 10 t ha ⁻¹	75.56	80.27	83.84	79.89	59.56	64.47	67.84	63.96	
M_2 : Green manure seed @ 25 kg ha ⁻¹	79.45	85.15	86.96	83.86	63.45	69.29	70.96	67.90	
M ₃ : FYM @ 5 t ha ⁻¹ + Green manures seed @ 12.5 kg ha ⁻¹	84.71	87.60	89.29	87.20	68.71	71.64	73.29	71.21	
M ₄ : FYM @ 10 t ha ⁻¹ + Azospirillum + PSB + KRB @ 1.25 l ha ⁻¹ each	92.52	93.33	94.14	93.33	80.52	81.60	82.14	81.42	
M ₅ : Green manure seed @ 25 kg ha ⁻¹ + $Azospirillum$ + PSB + KRB @ 1.25 l ha ⁻¹ each	92.90	97.93	100.83	97.22	80.90	85.93	87.50	84.78	
M ₆ : FYM @ 5 t ha ⁻¹ + Green Manures @ 12.5 kg ha ⁻¹ Azospirillum + PSB + KRB @ 1.25 l ha ⁻¹ each	95.81	101.74	102.12	99.89	83.81	88.40	90.12	87.44	
M ₇ : Control	54.05	68.30	74.98	65.78	36.38	51.63	56.98	48.33	
Mean	82.14	87.76	90.31		67.62	73.28	75.55		
	SEm +		CD (P=0.05)		SEm +		CD (P=0.05)		
М	2.30		7.09		2.33		7.18		
S	0.72		2.08		0.75		2.18		
S at M	1.90		5.99		1.99		6.19		
M at S	3.43		10.39		3.49		10.54		

Table 3: Effect of integrated nutrient management practices on acid and alkaline phosphatase activity ($\mu g \ p$ -nitrophenol g⁻¹ h⁻¹) at panicle initiationand harvest stage of paddy

Tractments	Acid phosph	atase	Alkaline phosphatase				
Treatments		U g - II -)	(µg <i>p</i> -mtrophene Deniele initiation				
Metrophete	ranicle initiatio	Inarvest	Panicie mitiatio	i narvest			
Main plots							
M ₁ : FYM @ 10 t ha ⁻¹	67.89	52.84	93.29	76.30			
M ₂ : Green manure seed @ 25 kg ha ⁻¹	71.00	56.02	97.33	80.37			
M ₃ : FYM @ 5 t ha ⁻¹ + Green manures seed @ 12.5 kg ha ⁻¹	76.10	61.08	102.68	86.67			
M ₄ : FYM @ 10 t ha ⁻¹ + Azospirillum + PSB + KRB @ 1.25 l ha ⁻¹ each	82.71	70.04	110.89	95.83			
M ₅ : Green manure seed @ 25 kg ha ⁻¹ + Azospirillum + PSB + KRB @ 1.25 l ha ⁻¹ each	85.44	72.48	114.00	99.01			
M ₆ : FYM @ 5 t ha ⁻¹ + Green manures @ 12.5 kg ha ⁻¹ + $Azospirillum$ + PSB + KRB @ 1.25 l ha ⁻¹ each	88.78	76.00	118.21	103.50			
M7: Control	53.33	36.02	75.70	58.96			
SEm_+_	2.20	2.28	3.24	3.03			
CD (P=0.05)	6.77	7.03	9.97	9.35			
Sub plots							
S1: 50% RDF	69.95	55.72	95.53	79.57			
S _{2:} 75% RDF	75.92	61.42	103.02	87.06			
S ₃ : 100% RDF	79.24	64.79	106.64	90.78			
SEm +	1.06	1.03	1.22	1.27			
CD (P=0.05)	3.08	2.99	3.54	3.68			
Interactions							
S at M							
SEm <u>+</u>	2.81	2.73	3.23	3.36			
CD (P=0.05)	NS	NS	NS	NS			

M at S				
SEm_+	3.51	3.59	4.94	4.71
CD (P=0.05)	NS	NS	NS	NS

Table 4: Effect of integrated nutrient management practices on microbial population at panicle initiation and harvest stage of paddy

Treatments		Bacteria (×10 ⁶ CFU g ⁻¹ soil)		Fungi (x10 ³ CFU g ⁻¹ soil)		Actinomycetes (x10 ⁴ CFU g ⁻¹ soil)		
		Harvest	Panicle initiation	Harvest	Panicle initiation	Harvest		
Main plots								
M ₁ : FYM @ 10 t ha ⁻¹	43.79	35.56	16.16	12.44	19.89	15.00		
M_2 : Green manure seed @ 25 kg ha ⁻¹	47.34	37.00	17.56	13.78	21.11	16.36		
M ₃ : FYM @ 5 t ha ⁻¹ + Green manures seed @ 12.5 kg ha ⁻¹	52.00	40.00	19.22	15.00	23.68	17.66		
M4: FYM @ 10 t ha ⁻¹ + Azospirillum + PSB + KRB @ 1.25 l ha ⁻¹ each	57.11	42.78	21.56	16.19	24.81	19.11		
M ₅ : Green manure seed @ 25 kg ha ⁻¹ + $Azospirillum$ + PSB + KRB @ 1.25 l ha ⁻¹ each	60.39	45.70	22.67	17.11	25.89	19.82		
M ₆ : FYM @ 5 t ha ⁻¹ + Green manures @ 12.5 kg ha ⁻¹ + $Azospirillum$ + PSB + KRB @ 1.25 l ha ⁻¹ each	63.46	48.33	24.22	18.56	28.44	21.64		
M7: Control	32.23	22.89	13.78	9.67	16.00	10.28		
SEm_+_	1.25	1.66	0.52	0.50	0.85	0.64		
CD (P=0.05)	3.86	5.12	1.59	1.55	2.63	1.97		
Sub plots								
S1: 50% RDF	46.71	35.57	17.54	13.05	21.10	15.25		
S _{2:} 75% RDF	51.52	39.62	19.52	14.81	22.82	17.47		
S _{3:} 100% RDF	54.47	41.49	20.86	16.18	24.59	18.66		
SEm_+_	0.86	0.56	0.37	0.32	0.35	0.37		
CD (P=0.05)	2.51	1.61	1.08	0.93	1.00	1.09		
Interactions								
S at M								
SEm <u>+</u>	2.29	1.47	0.99	0.85	0.92	0.98		
CD (P=0.05)	NS	NS	NS	NS	NS	NS		
M at S								
SEm <u>+</u>	2.21	2.50	0.93	0.86	1.32	1.07		
CD (P=0.05)	NS	NS	NS	NS	NS	NS		

Conclusion

From the results it can be concluded that among organic manures, at both the stages, the highest enzyme activity and microbial population was recorded with FYM @ 5 t ha⁻¹ + green manures @ 12.5 kg ha⁻¹ + *Azospirillum* + KRB @ 1.25 l ha⁻¹ each (M₆) which was on par with the application of green manures @ 25 kg ha⁻¹ + *Azospirillum* + KRB @ 1.25 l ha⁻¹ each (M₅) and FYM @ 10 t ha⁻¹ + *Azospirillum* + PSB + KRB @ 1.25 l ha⁻¹ each (M₄) and the lowest was recorded with control (M₇). However, among levels of fertilizers, the highest enzyme activity and microbial population was obtained with 100% RDF (S₃) which was comparable with 75% RDF (S₂) and 50% RDF (S₁).

Competing Interests

Authors have declared that no competing interests exist.

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