

Long term effect of fertilizers, manure and lime on biological health of an acid soil

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Received: September, 2021: Revised accepted: January, 2022

ABSTRACT

The study was conducted under long term fertilizer experiment (LTFE) to assess the impact of nutrient management practices on biological health of acid soils under soybean-wheat cropping system at Birsa Agricultural University, Kanke, Ranchi, Jharkhand. Ten treatments were evaluated in randomized block design with three replications. The integrated nutrient management involving the use of organic manure together with inorganic fertilization exerted a significant positive effect on soil microbial biomass carbon, CO₂ evolution, dehydrogenase activity (DHA), urease and microbial population. Similarly, use of lime along with NPK also showed improvement in enzymatic activities, soil microbial biomass carbon, and CO₂ evolution compared to 100% NPK and control. The microbiological properties were drastically reduced in 100% N only treatment. Application of 100% NPK was found to superior over 100% NP, 100% N and 50% NPK and 100% N(S)PK while the same was significantly at par with 150% NPK. The effect between weedicide applied and hand weeded treatments was also statistically at par for the grain and straw yield of wheat as well as for the biological properties of the soil. Long term application of FYM/lime with NPK significantly increased the mean grain and straw yield of wheat over the years (1972-2019) compared to only inorganic fertilization and control. Overall, continuous application of FYM or lime along with 100% NPK improved the soil biological health and increased the crop yield in acid soil under wheat based cropping system.

Key words: Soil biological health, acid soil, lime, FYM.

INTRODUCTION

Soil biological activity is one of the key factors determining the agricultural productivity and a sensitive indicator of anthropogenic influence. The biomass, metabolites, and microbial functions are directly related to soil fertility and many other soil properties that support agricultural production (Lehman *et al.* 2015) Physical fertility, chemical fertility, and biological fertility are three aspects of soil fertility that are all interconnected. Biological fertility is highly complex and dynamic and is the aspect that's the least understood. Soil microorganisms are of prime importance with regard to the biochemical processes occurring in soils like decomposition of organic matter, soil aggregate formation, soil humus formation nutrient transformation and nutrient cycling and play an essential role in maintaining soil productivity (Wu *et al.* 2011). Soil microorganisms are extremely sensitive to management practices such as the addition of mineral fertilizer and manure (Walsh *et al.* 2012). Therefore, soil biological properties have been used as the most subtle indicators of changes in the soil quality (Zornoza *et al.* 2015).

The biological properties of soil and their relationships with crops are more complicated, and fundamental knowledge is severely lacking (Lehman *et al.* 2015). Long term fertilizer experiments serve as an important tool to monitor the changes in the biological properties of soil as a basis for evaluating the soil quality and agricultural productivity. The present investigation was carried out to investigate the long term effect of various nutrient management options on biological properties of acid soil under continuous cropping.

MATERIALS AND METHODS

A long term fertilizer experiment was initiated during 1972-73 at Birsa Agricultural University, Kanke, Ranchi, Jharkhand, India (85° 19' E, 23° 17' N, 625 m above the mean sea level) in an acidic red loam soil under the All India Coordinated Research Project on Long - Term Fertilizer Experiments. The texture of the experimental soil was characterized as sandy clay loam and classified as Hyperthermic, mixed Typic Paleustalf. The climate of the experimental site is sub-tropical. Total annual precipitation is

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about 1400 mm. The experiment started with soybean-potato-wheat cropping system (1972-1979) which was changed to soybean-toria-wheat (1980-1985) and soybean-wheat cropping system (since 1986), respectively. The experiment was laid out in randomized block design having 100 m² individual plot size and consisted of ten treatments: T₁ (50% NPK + Weedicide), T₂ (100% NPK + weedicide), T₃ (150% NPK+Weedicide), T₄ (100% NPK + Hand weeding), T₅ (100% NPK+lime+weedicide), T₆ (100% NP + weedicide), T₇ (100% N + weedicide), T₈ (100% NPK + FYM + weedicide), T₉ (100% N(S)PK +weedicide) and T₁₀ (control). Based upon initial soil test value, the fertilizer dose for soybean and wheat was 25: 60:40 kg ha⁻¹ (N: P₂O₅: K₂O) + *Rhizobium* culture and 80:60:40 kg ha⁻¹ (N: P₂O₅: K₂O), respectively. Considering the nitrogen requirement and phosphorus accumulation in the P treated plots, the fertilizer dose was revised to 25:30:40 and 120:30:40 N: P₂O₅:K₂O kg ha⁻¹, for soybean and wheat, respectively. The source of N, P and K fertilizer was urea, diammonium phosphate (DAP) and muriate of potash (MOP) in all treatments except in T₉ where source of P was single super phosphate (SSP). In T₉, initially the source of N was ammonium sulphate which has been replaced by urea from the year 2010-11. In T₅, lime is applied @1/10 of LR to *kharif* crop. In T₈, FYM is applied@ 10 t ha⁻¹ on fresh weight basis to *kharif* crop. Imazethapyr as pre-emergence weedicide and 2,4-D as post emergence weedicide was applied in soybean and wheat crop in all the treatments except 100% NPK+HW and control. The original experimental set up is having 4 replications, but for the present study 3 replications were considered.

Surface soil samples were collected after the harvest of wheat during 2018-19 Samples were processed and analyzed for soil pH, soil organic carbon and different biological properties. Soil microbial biomass carbon (SMBC) was measured by chloroform fumigation extraction method as described by Jenkinson and Powlson (1976). CO₂ evolution was determined by alkali trap method described in Chhonkar *et al.* (2007). Dehydrogenase activity (DHA) in soil was estimated by extracting the soil with 2,3,5 triphenyl tetrazolium chloride (TTC) (Klein *et al.* 1971) which was reduced to triphenyl formazan (TPF). Soil urease activity

was determined by the method described in Chhonkar *et al.* (2007). The method for assay of urease activity in soil involved the estimation of urea hydrolysis in soil by determination of urea remaining after the incubation of soil with urea solution. The difference between the amount of urea added and that recovered after incubation for specific time was taken as an estimate of urease activity. The total number of bacteria, actinomycetes and fungi were determined as colony forming units using respective media through serial dilution methods (Wollum 1982).

RESULTS AND DISCUSSION

Soil reaction, soil organic carbon and wheat grain and straw yield

After 48 years of long term application of fertilizers, manure and lime soil pH ranged from 4.3 in 100% N to 5.9 in 100% NPK + Lime treatment (Table1). Continuous application of lime along with balanced chemical fertilizers increased the soil pH by 13.46 % and 7.2% over T₂ (100%NPK+Weedicide) and control, respectively. In general, there was a decline in soil pH due to continuous addition of only inorganic fertilizers without lime/FYM compared to that of control. Continuous application of 100% N alone resulted in drastic reduction in soil pH. The acid producing nature of these inorganic fertilizers may be the reason for the decline in soil pH. Soil pH in T₉ (100%N(S) PK + Weedicide) was low as compared to T₂ (100%NPK+Weedicide) or T₄ (100% NPK + Handweeding) which may be due to use of ammonium sulphate as source of N since start of the experiment till 2010. Fageria *et al.* (2010) also reported that decrease in soil pH was greater when ammonium sulphate was used as the N source compared to urea. Continuous use of lime /FYM in acid soil improved the soil pH as compared to control and other inorganic treatments. Soil organic carbon ranged from the lowest value of 3.9 g kg⁻¹ in 100% N alone treatment to the highest value of 5.9 g kg⁻¹ in 100% NPK+FYM treatment (Table1). Continuous addition of FYM along with 100% NPK resulted in significantly higher organic carbon accumulation as compared to rest of the treatments. The enhanced soil organic carbon in FYM treated plot might be due to regular addition of organic materials for forty eight years

and also due to greater crop productivity and hence return of more crop residues. Increase in SOC with addition of organic manure has been reported by many workers (Bhattacharyya *et al.* 2011). Application of only NPK fertilizers (T₁, T₂, T₃, T₄, T₉) as well as along with lime (T₅) also

showed improvement in SOC concentration over control. Plant root and shoot biomass increased with application of fertilizers in these treatments leading to addition of organic residues in soil (Gathala *et al.* 2007).

Table 1: Effect of continuous use of fertilizer, manure and lime on soil pH, soil organic carbon (SOC) and mean grain and straw yield of wheat

Treatment Details	Soil pH	SOC (g kg ⁻¹)	Mean grain yield (q ha ⁻¹) (1972-2019)	Mean straw yield (q ha ⁻¹) (1972-2019)
T ₁ : 50%NPK+W	5.3	4.3	20.53	33.09
T ₂ : 100%NPK+W	5.2	4.8	28.68	46.05
T ₃ : 150%NPK+W	5.1	4.6	29.52	47.8
T ₄ : 100%NPK+HW	5.2	5.0	29.12	48.45
T ₅ : 100%NPK+Lime+W	5.9	4.9	32.48	52.05
T ₆ : 100%NP+W	5.2	4.0	26.16	37.99
T ₇ : 100%N+W	4.3	3.9	4.75	10.08
T ₈ : 100%NPK+FYM+W	5.7	5.9	34.56	54.36
T ₉ : 100%N(S)PK+W	4.8	4.6	20.27	32.58
T ₁₀ : Control	5.5	4.1	6.67	11.14
CD (P=0.05)	0.19	0.82	4.45	7.51

W: Weedicides, HW: Handweeding

After forty eight years of continuous cropping mean grain yield of wheat varied from 4.75 q ha⁻¹ to 34.56 q ha⁻¹ and mean straw yield varied from 10.28 q ha⁻¹ to 54.36 q ha⁻¹ in 100% N and 100% NPK+FYM, respectively (Table 1). Grain yield of wheat in plots receiving FYM along with balanced NPK fertilizers was significantly high as compared to treatments involving only chemical fertilizers and control and at par with 100% NPK+Lime. 100% NPK+FYM also recorded the highest straw yield followed by 100%NPK+Lime (Table1). Higher accumulation of organic carbon due to continuous application of FYM over 48 years contributed to higher crop yield. Positive impact of lime on soil physical properties is related to its effect on increasing crop growth, soil organic matter content and thus improved soil biological activity (Manna *et al.* 2007). Imbalanced application of inorganic fertilizers (100% NP and 100% N), 50% NPK and 100%N(S)PK resulted in significant decrease in grain and straw yield as compared to 100% NPK and 150%NPK. Drastic reduction was noticed in 100% N where the productivity was even lower than the control treatment. The mean grain yield of wheat (20.27 q ha⁻¹) and straw yield (32.58 q ha⁻¹) was observed to be low in 100%N(S) PK when compared to 100%NPK which may be attributed to lower yield of wheat during initial years which may be due to the use of ammonium sulphate as N source which led to

reduced soil pH. The mean grain and straw yield of wheat in 150% NPK+Weedicide and in the treatment consisting of handweeding (100%NPK+HW) were similar to treatment using the chemical methods of weed control and 100%NPK (i.e. T₂ 100% NPK+Weedicide) and did not differ significantly. It was noticed that increasing the dose of inorganic fertilizers from suboptimal to optimal resulted in significant increase in the grain as well as the straw yield.

Soil microbial biomass carbon (SMBC)

Soil microbial biomass carbon was maximum in treatment 100% NPK+FYM (307.74 µg g⁻¹ soil) followed by 100% NPK+Lime (292.06 µg g⁻¹ soil) (Table 2). Inclusion of FYM and lime along with 100%NPK registered an increase of 23.30% and 17.0%, respectively, over T₂ (100%NPK+Weedicide). Continuous application of organic manure in conjunction with 100% NPK significantly influenced the SMBC content as compared to rest of the treatments which may be ascribed to the supply of additional mineralizable and readily hydrolysable carbon due to organic manure addition which enhanced the microbial activity and hence SMBC (Mali *et al.*, 2015). Application of lime along with balanced

inorganic fertilizers had a positive impact on SMBC as lime has an ameliorative effect on soil pH improving the soil condition for microbial activity and proliferation. The treatments T₂, T₃ and T₄ was statistically superior to the treatments receiving imbalanced fertilization (100% NP and 100 % N), 50% NPK, 100% N(S)PK and control . Restricted crop production and hence carbon substrate in control, imbalanced treatments, 50% NPK due to greater stress due to no or inadequate use of nutrients resulted in lower SMBC as compared to 100% NPK and 150% NPK treatments. Increasing dose of chemical fertilizers from 50% to 100% NPK increased the SMBC due to formation of more root exudates and underground root residues (Bedi *et al.* 2009). Application of super-optimal dose of chemical fertilizers did not differ significantly over optimal dose. SMBC in treatment involving hand weeding was at par with that of chemical method of weed control. Continuous application of N alone had the most deleterious effect on SMBC. The lowest value of SMBC in 100%N plot may be due to unfavourable environment for soil microorganisms on account of low pH due to use of urea alone over the years coupled with low crop productivity and hence low organic carbon content. Verma *et al.* (2017) also observed that application of FYM or lime along with NPK fertilizer had significant effect on SMBC content in acid soil.

CO₂ Evolution

Significant influence of FYM or lime in conjunction with 100% NPK fertilizers was observed on soil respiration over chemical fertilizers alone and control (Table 2). Increased microbial biomass in FYM and lime treated plot could have resulted in higher soil respiration. Enhanced microbial activity in lime treated plot on account of favourable pH, higher root biomass due to increased productivity may have resulted in greater CO₂ evolution as compared to only NPK treatments. Kumari *et al.* (2019) also reported higher CO₂ evolution due to application of FYM or lime in conjunction with chemical fertilizers in acid soil. All the treatments were significantly higher than control and 100% N. It was observed that application of 100% NPK with or without chemical weedicides (T₂ and T₄) and 150% NPK showed significant improvement in CO₂ evolution as compared to imbalanced fertilization (100% N, 100% NP), suboptimal dose of chemical fertilizers (50%NPK) and 100%N(S)PK. Higher accumulation of root biomass associated with greater yield in 100% NPK and 150% NPK be the reason of increased CO₂ evolution. Lower CO₂ evolution was recorded in 100%N(S)PK as compared to 100% NPK which might be associated with lower soil pH and higher salt content in 100%N(S)PK treatment due to the use of ammonium sulphate during previous years which might hamper the microbial activity. The lowest value of SMBC was recorded in 100% N treatment which may be due to unfavourable environment for soil microorganisms on account of low pH due to use of urea alone over the years.

Table 2: Effect of continuous use of fertilizer, manure and lime on soil microbial biomass carbon (SMBC), CO₂ evolution, dehydrogenase activity (DHA) and urease activity

Treatment Details	SMBC ($\mu\text{g g}^{-1}$ soil)	CO ₂ evolution (mg 100 g ⁻¹ soil)	DHA ($\mu\text{g TPF g}^{-1}$ soil 24 h ⁻¹)	Urease ($\mu\text{g urea-N hydrolyzed}$ g ⁻¹ soil hr ⁻¹)
T ₁ : 50%NPK+W	190.69	54.08	27.08	14.87
T ₂ : 100%NPK+W	249.57	66.00	33.75	19.02
T ₃ : 150%NPK+W	252.77	63.25	31.69	17.32
T ₄ : 100%NPK+HW	253.91	67.83	33.62	18.72
T ₅ : 100%NPK+Lime+W	292.06	77.28	39.21	22.41
T ₆ : 100%NP+W	158.66	51.33	24.91	12.56
T ₇ : 100%N+W	121.96	34.65	10.67	5.70
T ₈ : 100%NPK+FYM+W	307.74	78.74	41.52	26.04
T ₉ : 100%N(S)PK+W	200.60	51.43	20.44	10.68
T ₁₀ : Control	140.85	40.88	14.20	9.07
CD (P=0.05)	12.76	9.18	3.14	2.31

Dehydrogenase activity (DHA)

The dehydrogenase activity (DHA) showed significant improvement with the addition of lime or FYM along with 100%NPK over only inorganic fertilizers. The maximum amount of DHA was recorded in 100%NPK+FYM treatment ($41.52 \mu\text{g TPF g}^{-1} \text{ soil } 24 \text{ h}^{-1}$) which was statistically at par with 100%NPK+Lime ($39.21 \mu\text{g TPF g}^{-1} \text{ soil } 24 \text{ h}^{-1}$) (Table 2). Continuous application of FYM and lime together with 100% NPK for forty eight years enhanced the DHA by 23% and 16.2%, respectively, over T_2 (100% NPK+Weedicide). Greater amount of organic substrate in FYM treated plots might have resulted in high microbial activity which results in high dehydrogenase activity (Basak *et al.* 2013). Choudhary *et al.* (2021) also observed that application of organic manure along with inorganics increased the DHA significantly. Increase in population of bacteria due to increased soil pH by continuous application of lime may have resulted in higher DHA in lime treated plots (Vishwanath *et al.*, 2020). Continuous application of 150% NPK and practice of hand weeding did not differ significantly either over 100% NPK or chemical control of weeds. It was observed that continuous application of imbalanced fertilization (100% N and 100% NP) decreased the DHA by 68.38% and 26.19% over T_2 (100%NPK+Weedicide). The decrease was most spectacular in 100%N alone where DHA was significantly lower than the control. Vishwanath *et al.* (2020) also recorded significantly lowest DHA in 100%N treatment which could be due to increased redox potential of soil owing to accumulation of nitrate and other anions due to continuous application of N. Increasing dose of chemical fertilizers from suboptimal (50%NPK) to optimum (100% NPK) significantly increased the DHA which may be due to the fact that microbial activity was more stimulated due to balanced fertilization to utilize the native pool of organic carbon as a source of carbon, which acts as substrate for dehydrogenase (Bedi *et al.* 2009). 100%N(S)PK treatment was also significantly low in DHA as compared to 100%NPK which might be attributed to lower soil pH. Activities of beneficial microorganisms, except fungi, are restricted due to soil acidity.

Soil urease activity

Soil urease activity ranged from the lowest value of $5.70 \mu\text{g urea-N hydrolyzed g}^{-1} \text{ soil hr}^{-1}$ in 100%N to the highest value of $26.04 \mu\text{g urea-N hydrolyzed g}^{-1} \text{ soil hr}^{-1}$ in 100%NPK +FYM treatment (Table 2). 100%NPK+FYM recorded significantly higher urease activity as compared to rest of the treatments. Maximum increase in urease activity with continuous addition of FYM along with NPK is because addition of organic sources acts as good source of carbon and energy to heterotrophs by which their population increases with increase in soil enzyme activity (Rai and Yadav 2011). Addition of lime along with balanced NPK fertilizers provided suitable environment for microbial activity by exerting ameliorative effect on soil pH suitable for soil microbes and inorganic fertilizers provided easily accessible nutrient for microbes. It was observed that increase in fertilizer level from suboptimum to optimum level significantly increased urease activity but further increase to super optimal dose did not differ significantly over 100%NPK. In general, application of balanced chemical fertilizers stimulated the growth and multiplication of microorganisms as compared to suboptimal and imbalanced fertilization. T_2 , T_3 and T_4 were found to be at par. 100% N(S) PK treatment recorded lower value of urease activity when compared to 100% NPK treatments (T_2 and T_4) which could be attributed to low soil pH and higher salt concentration T_2 and T_4 making the soil environment not so conducive for microbial actions. Lowest urease activity associated with control and 100% N could be due to insufficient substrate on concentration of enzyme i.e. organic carbon which acts as an energy source for microbial population (Kanchikerimatha and Singh, 2001).

Microbial Population

The population of bacteria and actinomycetes was significantly influenced by the application of lime or FYM along with 100%NPK treatment over rest of the treatments (Table 3.). Higher count of bacteria and actinomycetes in lime and FYM treated plots ($7.47 \log_{10} \text{ CFU}$ and $7.52 \log_{10} \text{ CFU}$ in 100% NPK+Lime and 100% NPK + FYM, respectively for bacteria ; $6.19 \log_{10} \text{ CFU}$ and $6.17 \log_{10} \text{ CFU}$

in 100% NPK + FYM and 100% NPK + Lime, respectively for actinomycetes) could be attributed to favourable effect on soil reaction through lime which supports higher microbial growth and to the large amount of readily available carbon from FYM that acts as a substrate for stimulation of microorganisms. Application of lime favours neutral pH and add Ca which is conducive for bacterial growth. Moreover addition of organic matter improve soil physical environment making it more suitable for proliferation of soil microorganisms (Tejada *et al.* 2008). Continuous application of only inorganic fertilizers recorded significantly lower count of bacteria and actinomycetes compared to integrated use of manure or lime and fertilizers suggesting the importance of organics and lime in improving the biological health of acid soil. Significant increase in bacteria and actinomycetes in 100%NPK treatment as compared to 50%NPK treatment and imbalanced fertilization was observed which could be due to higher root biomass due to higher crop productivity with balanced fertilization which in turn provided greater

substrate for source of energy for growth of actinomycetes and bacteria. Population of actinomycetes and bacteria was considerably low in 100% N alone due to low soil pH and inadequate carbon and nutrient sources. Gowda *et al.* (2017) also observed that application of lime and/or FYM supports higher population of bacteria and actinomycetes. In contrast to bacteria, integrated application of lime along with NPK recorded significantly low population of fungi which may be due to higher soil pH in lime treated plot. Higher fungal population was observed in 100% N and 100% N(S)PK plot unlike bacteria or actinomycetes due to low soil pH which is preferred by the fungi (Saha *et al.* 2008). Vishwanath *et al.* (2020) also reported increased population of fungi in 100% N plot as compared to 100% NPK+Lime treatment in acidic Alfisol. Increasing level of chemical fertilizer from 50% NPK to 150% NPK registered significant increase in fungal population due to increase in the carbon substrate resulting from higher amount of decomposed organic matter due to increased root biomass.

Table 3: Effect of continuous use of fertilizer, manure and lime on soil microbial population [\log_{10} colony forming units (CFU)]

Treatment Details	Bacteria	Actinomycetes	Fungi
T ₁ : 50%NPK+W	7.26	6.03	4.00
T ₂ : 100%NPK+W	7.36	6.10	4.09
T ₃ : 150%NPK+W	7.32	6.05	4.11
T ₄ : 100%NPK+HW	7.40	6.10	4.06
T ₅ : 100%NPK+Lime+W	7.47	6.17	3.91
T ₆ : 100%NP+W	7.20	5.97	4.01
T ₇ : 100%N+W	6.97	5.68	4.04
T ₈ : 100%NPK+FYM+W	7.52	6.19	4.20
T ₉ : 100%N(S)PK+W	7.12	5.95	4.17
T ₁₀ : Control	7.20	5.92	3.82
CD (P=0.05)	0.06	0.06	0.06

Continuous use of urea only as N source reported the highest decline in the grain and straw yield of wheat crop and had deleterious effect on microbiological properties of acid soil. The superiority of balanced fertilization in maintaining the crop productivity and biological health of acid soil under continuous cultivation over imbalanced fertilization was clearly observed. Practice of manual method of weed control was at par with chemical control with respect to productivity as well as biological properties. Similarly, application of super-optimal dose of chemical fertilizers was also at par with 100%NPK. The study also showed that

use of ammonium sulphate as N source along with P and K fertilizer led to significant deterioration in the biological properties of soil as compared to use of urea as N source together with P and K sources. Further, significant improvement in grain and straw productivity of wheat crop and biological properties of soil was noticed with the integrated application of FYM or lime along with balanced NPK. Thus, under acidic environment use of FYM or lime in conjunction with balanced inorganic fertilizers must be advocated for ensuring higher agricultural productivity and improving the biological health of soil.

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