



Soil Properties as Influenced by Long Term Herbicide with or without Organic Manure Application in Transplanted Rice-Rice System

K. Goswami, S. Dutta, N. C. Deka, K. Das, D. K. Patgiri, H. Uddin and N. Borah*

Department of Soil Science, Assam Agricultural University,
Jorhat-785013, Assam, India

Abstract: Transplanted autumn rice-winter rice is one of the major rice-based cropping systems followed under specific soil type and land situation that faces diverse weed flora and intensity. The soil physical, chemical and biological properties of a long-term trial, being conducted since 2001 at Assam Agricultural University, Jorhat, India were evaluated after completion of tenth crop cycles of rice-rice cropping sequence. Rotational application of pre-emergence, followed by post-emergence herbicide significantly increased grain yield in both the crops. The pH of the soil increased and fractions of soil acidity decreased significantly, while the cation exchange capacity and exchangeable Ca^{2+} , Mg^{2+} and NH_4^+ increased significantly by organic manure application for partial N-fertilizer substitution. The application of organic manure significantly increased organic carbon and positively influenced available nitrogen and phosphorous content in soil. The microbial biomass carbon, urease activity were unaffected by herbicide application but were enhanced by organic manure addition. Application of herbicide significantly reduced the acid phosphatase activity in soil, while it had a positive effect on dehydrogenase activity and the effect was pronounced by addition of the organic manure. The effect of rotation of herbicide was not obvious and was limited to a few biological properties.

Key words: Long-term use, herbicide, organic manure, transplanted rice, soil properties

Introduction

Rice (*Oryza sativa* L.) occupies about two-third of the total cropped area in Assam and autumn rice-winter rice is one of the major rice-based cropping systems followed in specific soil type and land situations. The use of organic manure along with chemical fertilizers directly and indirectly affects soil properties (Nayak *et al.* 2017) yield and quality of crop (Shikhu *et al.* 2017). The long term effects of organic and chemical fertilizer on soil properties and nutrient status had been reported (Mandal *et al.* 2007). The cultivation of rice mainly as rainfed or in puddled and

continuous flooded soil condition, weed menace is one of the major constraints to higher productivity. Uncontrolled weeds may cause a reduction of grain yield up to 45 per cent in transplanted rice (Singh *et al.* 2004), which faces diverse weed flora and intensity due to alternate wetting-drying cycles throughout the crop growth period. Application of herbicides had been reported to increase 30 to 40 per cent yield over control in transplanted rice (Bari 2010). Many biological and biochemical parameters are sensitive to long term application of herbicides as mixture or in sequence to control broad spectrum of weeds in rice. The application of butachlor was shown to affect soil enzyme activity

*Corresponding author: (Email: nilayborah@rediffmail.com)

differently under upland (Latha and Gopal 2010) and flooded conditions (Rasool *et al.* 2014). Long term use of herbicides significantly influence the soil microbial community population and activity (Abbas *et al.* 2015), which have strong influences on the processes such as decomposition of organic matter and nutrient cycling in the soil (Zeller *et al.* 1995). Beneficial effects of long term farmyard manure addition with butachlor and 2, 4-D in rice-rice system had been reported in terms of faster degradation of the herbicides and facilitating soil microbial population in soil (Devi *et al.* 2007). However, the effects may vary considerably with quality of organic manure (Mohanty *et al.* 2013), soil type and climate. Accordingly the present work was carried out to evaluate soil properties in a long-term experiment of weed and nutrient management in rice-rice cropping sequence.

Materials and Methods

The study was carried out in the long-term field experiment that had been continued since April, 2001 at the Instructional-Cum-Research (ICR) Farm, Assam Agricultural University, Jorhat, situated at 26°47' N latitude, 94°12' E longitude and at an altitude of 86.6 m from the mean sea level. The climatic condition of Jorhat is sub-tropical and humid having hot summer and cold winter with average annual rainfall is 2042 mm, maximum temperature 34°C to 47°C during summer and the minimum temperature between 8 to 10°C during winter. Soil samples were collected from the respective plots of the following treatments:

T₁ - Hand weeding once in each crop with recommended dose of fertilizer (RDF) 40:20:20 and 60:20:40 kg ha⁻¹ N:P₂O₅:K₂O in autumn rice and winter rice, respectively.

T₂ - Butachlor 1.0 kg ha⁻¹a.i. followed by 2,4-D 0.75 kg ha⁻¹a.i. with RDF in each crop

T₃ - Butachlor 1.0 kg ha⁻¹a.i. followed by 2,4-D 0.75 kg ha⁻¹a.i. with RDF (25 % N substitution through organic manure)

T₄ - Butachlor 1.0 kg ha⁻¹a.i. in autumn and pretilachlor 0.75 kg ha⁻¹a.i. in winter rice followed by 2, 4-D 0.75 kg ha⁻¹a.i. in each crop with RDF

T₅ - Butachlor 1.0 kg ha⁻¹a.i. in autumn and pretilachlor 0.75 kg ha⁻¹a.i. in winter rice followed by 2, 4-D 0.75 kg ha⁻¹a.i. Butachlor 1.0 kg ha⁻¹a.i. in autumn and pretilachlor 0.75 kg ha⁻¹a.i. in winter rice followed by 2,4-D 0.75 kg ha⁻¹a.i. in each crop with RDF with RDF (25 % N substitution through organic manure).

The soil of the experimental site belongs to Aeric Endoaquept with sandy clay loam texture (Dutta and Karmakar 1995) and had pH 5.0, available N 243.5 kg ha⁻¹, P₂O₅ 18.1 kg ha⁻¹ and K₂O 93.6 kg ha⁻¹. The experiment was laid out in completely Randomized Block Design (RBD) with six replications and individual plot size of 50 m². Representative surface soil (0-15 cm) sample from each plot was collected after harvest of winter rice in December (20th and 22nd crop) and autumn rice in July (21st crop) for analysis of available nutrients in soil. The soil sample collected after the 20th crop was also used for analysis of chemical properties, microbial population and other biological properties. Soil pH was determined by using glass electrode pH meter (Jackson 1973). Cation exchange capacity and total acidity was determined by using method described by Baruah and Barthakur (1997). Exchangeable Ca²⁺ and Mg²⁺ was determination by Versenate titration method (Richards 1954) and determination of exchangeable Na⁺ and K⁺ flame-photometrically (Jackson 1973). Extraction of exchangeable Al was obtained by leaching the soil with 1 N KCl solution (Hesse 1971) and Al³⁺ in the extract was estimated colorometrically by the method described by Sivasubramaniam and Talibudeen (1972). The soil was extracted with 1 N Na₂SO₄-phenyl mercuric acetate and the exchangeable NH₄⁺ in the solution was estimated calorimetrically (Onken and Sunderman 1977). Exchange acidity of the soils was estimated using 1 N KCl solution as extractant and the extract was titrated with 0.1 N NaOH as described by Baruah and Barthakur (1997). The total acidity in soils was estimated by the method described by Kappen (1934) using 1 N NaOH for extraction and titrated with 0.1 N NaOH solution. Organic carbon content was determined by Walkley and Black wet digestion method as described by Baruah and Barthakur (1997). Available nitrogen in soil was

determined by alkaline potassium permanganate method (Subbiah and Asija 1956). Available phosphorous in soil was determined by Bray and Kurtz No 1 method (Jackson 1973). Available potassium in soil was determined by extracting the soil with neutral normal ammonium acetate and the potassium in the extract determined flame-photometrically as outlined by Jackson (1973). Serial dilution technique was used for isolation of *Azotobacter*, *Azospirillum* and PSB in appropriate media. The soil microbial biomass carbon was estimated through Fumigation Extraction method described by Jenkinson and Powlson (1976). The urease activity in soil was estimated by method described by Tabatabai and Bremner (1969). Acid phosphatase activity was detected colorimetrically following the method of Tabatabai and Bremner (1969). Dehydrogenase activity was determined by reduction of triphenyltetrazolium chloride (TTC) to triphenylformazone (TPF) as described by Casida (1968). A one-way ANOVA was carried out to compare the means of the different treatments. When significant F-values were detected, the differences between

individual means were tested using the critical difference (CD) test at 5 % probability level.

Results and Discussion

Grain Yield

The grain yield of rice is presented in table 1. The highest grain yield of 2860 kg ha⁻¹ in autumn rice was recorded with rotation of pre-emergence herbicide with RDF. In case of winter rice, rotation of pre-emergence herbicide and addition of organic manure for substitution of N-fertilizer in RDF had the highest yield of 4530 kg ha⁻¹. Continuous use of same herbicides had no effect on grain yield of autumn rice as compared to one hand weeding in each crop. Irrespective of the crops, partial substitution of N-fertilizer through organic manure was at par with sole chemical fertilizer. Partial substitution of N-fertilizer through organic manure did not have any effect on grain yield. Significant increase on grain yield of rice with substitution of recommended dose of fertilizers through organic manure in short-term experiments had been reported Rama Lakshmi *et al.* (2012).

Table 1. Grain yield of rice (kg ha⁻¹) as affected by treatments

Treatment	Autumn rice (21 st crop)	Winter rice (22 nd crop)
Hand weeding once in each crop with RDF	2310	3765
Butachlor followed by 2,4-D in each crop with RDF	2725	4380
Butachlor followed by 2,4-D in each crop with RDF (25 % N substitution through organic manure)	2640	4475
Butachlor (autumn) or pretilachlor (winter) followed by 2,4-D in each crop with RDF	2860	4505
Butachlor (autumn) and pretilachlor (winter) followed by 2,4-D in each crop with RDF (25 % N substitution through organic manure)	2785	4530
CD _{P=0.05}	420	370
CV (%)	6.7	5.6

RDF= 40:20:20 and 60:20:40 kg ha⁻¹ N:P₂O₅:K₂O in autumn rice and winter rice, respectively

Soil pH and soil acidity, exchangeable aluminum and base saturation

The pH of the soil was highly acidic pH (4.86-5.02). The pH of the soil significantly increased due to organic manure addition, while application of herbicides did not have any effect on soil pH (Table 2). Application of herbicides showed no significant effect on total

acidity, exchange acidity and exchangeable Al. The base saturation of the soil was unaffected by the treatments. The increase in soil pH even after long-term use of organic manure was low which might be due to low application rate of the manure and buffering capacity of the soil (Boateng *et al.* 2006).

Table 2. Soil pH, total acidity, exchange acidity, exchangeable aluminum [c mol (p⁺) kg⁻¹] and base saturation (%) after harvest of 20th crop

Treatment	pH	Total acidity	Exchange acidity	Exchangeable Al ³⁺	Base saturation (%)
Hand weeding once in each crop with RDF	4.88	1.92	0.69	1.25	37.0
Butachlor followed by 2,4-D in each crop with RDF	4.89	1.97	0.67	1.26	35.6
Butachlor followed by 2,4-D in each crop with RDF (25 % N substitution through organic manure)	5.02	1.81	0.62	1.11	38.5
Butachlor (autumn) or pretilachlor (winter) followed by 2,4-D in each crop with RDF	4.89	1.93	0.70	1.24	35.9
Butachlor (autumn) and pretilachlor (winter) followed by 2,4-D in each crop with RDF (25 % N substitution through organic manure)	5.01	1.79	0.59	1.12	38.7
CD _{P=0.05}	0.11	0.13	0.07	0.11	-
CV (%)	1.89	5.8	8.5	7.3	-

RDF= 40:20:20 and 60:20:40 kg ha⁻¹ N:P₂O₅:K₂O in autumn rice and winter rice, respectively

Table 3. Cation exchange capacity (CEC) and exchangeable cations [c mol (p⁺) kg⁻¹] after harvest of 20th crop

Treatment	CEC [c mol (p ⁺) kg ⁻¹]	Exchangeable cations [c mol (p ⁺) kg ⁻¹]			
		K ⁺	NH ₄ ⁺	Na ⁺	Mg ²⁺
Hand weeding once in each crop with RDF	5.92	0.05	0.24	0.12	1.14
Butachlor followed by 2,4-D in each crop with RDF	5.99	0.05	0.24	0.11	1.16
Butachlor followed by 2,4-D in each crop with RDF (25 % N substitution through organic manure)	6.21	0.06	0.25	0.11	1.28
Butachlor (autumn) or pretilachlor (winter) followed by 2,4-D in each crop with RDF	5.97	0.05	0.24	0.13	1.12
Butachlor (autumn) and pretilachlor (winter) followed by 2,4-D in each crop with RDF	6.27	0.06	0.25	0.11	1.29
(25 % N substitution through organic manure)	0.22	NS	0.01	NS	0.11
CD _{P=0.05}	3.0	9.8	4.7	9.9	7.8
CV (%)					8.7

RDF= 40:20:20 and 60:20:40 kg ha⁻¹ N: P₂O₅: K₂O in autumn rice and winter rice, respectively.

Cation exchange capacity (CEC) and exchangeable cations

The cation exchange capacity of the soil was significantly increased due to organic manure addition for partial N-fertilizer substitution, compared to chemical fertilizer alone (Table 3). The CEC ranged from 5.92 to 6.27 cmol (p⁺) kg⁻¹, the highest value of 6.27 cmol (p⁺) kg⁻¹ being recorded in soil receiving rotation of pre-emergence herbicide with 25 % N-fertilizer substitution through organic manure. The exchangeable cations *viz.* NH₄⁺, Ca²⁺ and Mg²⁺ significantly increased due to organic manure application, while the difference among the treatments was not significant in case of K⁺ and Na⁺. The increase in exchangeable Ca²⁺ and Mg²⁺ in soil, may be ascribed to their relative contents in the organic manure, to the line of 1.0–1.2 % Ca and 0.3–0.5 % Mg. The effect on exchangeable K⁺ may be attributed to relatively high demand for K by both the crops, while the same for exchangeable Na might be due to relatively lower preference of soil exchange sites for Na⁺.

Soil organic carbon and available nutrient status in soil

The organic carbon content significantly increased due to rotation of pre-emergence herbicide and addition of organic manure for substitution of N-fertilizer in winter rice (9.4 g kg⁻¹ after 20th and 22nd crop) and in autumn rice (9.8 g kg⁻¹ after 21st crop), but was at par with organic manure addition without pre-emergence herbicide rotation (Table 4). The effect of weed management practices on soil organic carbon content was statistically non-significant. The treatment receiving organic manure for partial N-fertilizer substitution without herbicide rotation also showed significantly higher available nitrogen content with sole chemical fertilizer after harvest of autumn rice (21st crop), while after harvest of winter rice (22nd crop) the significant difference was observed only with sole chemical fertilizer with hand weeding. In general, relatively lower content of available nitrogen in soil was recorded after harvest of autumn rice than winter rice. The P₂O₅ content in soil after harvest of winter rice (20th and 22nd crop) was statistically higher in soil receiving herbicide rotation and organic manure over sole chemical fertilizer, but was at par with organic manure

for partial N-fertilizer substitution without herbicide rotation. Addition of organic manure for partial N-fertilizer substitution without herbicide rotation resulted significantly higher P₂O₅ content in soil only in 20th crop, after harvest of winter rice. Irrespective of the seasons and years, the available potassium (K₂O) content in soil was not affected by the treatments, and no definite trend was observed. The effect of long-term application of organic manure on available potassium was not conspicuous. This might be due to relatively high demand and uptake for potassium in both the crops. Application of organic manure with and without chemical fertilizers had been reported to increase organic carbon and available nutrients over sole chemical fertilizer (Srinivasarao *et al.* 2012).

Soil microbial properties

The count of *Azotobacter*, *Azospirillum* and PSB were significantly higher in the plots receiving hand weeding once in each crop, and plots receiving herbicides with organic manure for partial N-fertilizer substitution compared to the soils treated with herbicide without organic manure (Table 5). However, the highest values, *i.e.* 17.4, 12.6 and 17.7 (-log x 10⁶ cfu g⁻¹ soil), were observed in hand weeding plot for *Azotobacter*, *Azospirillum* and PSB, respectively. However, addition of organic manure for partial N-fertilizer substitution significantly increased microbial biomass carbon in soil. The highest value of 370.1 µg g⁻¹ soil was recorded with rotation of herbicide and organic manure addition for partial N-fertilizer substitution.

Enzyme activity

The acid phosphatase in soil was significantly affected by application of herbicides compared to hand weeding and the highest value was 200.4 µg p-nitrophenol g⁻¹ dry soil h⁻¹ (Table 6). The activity of the enzyme in soils was significantly lower with the application of chemical fertilizer alone than those with organic manure. Application of butachlor did not change the urease activity despite an initial increase during first ninety days after application. However, among the herbicide-treated soils, the increase in urease activity with organic manure addition may be due to the positive effect of organic manure in soil. Application of

Table 4. Organic carbon content (g kg^{-1}) available N, P_2O_5 and K_2O (kg ha^{-1}) in soil after harvest of 20th, 21st and 22nd crop

Treatments	Organic carbon			Available-N			Available- P_2O_5			Available- K_2O		
	20 th	21 st	22 nd	20 th	21 st	22 nd	20 th	21 st	22 nd	20 th	21 st	22 nd
Hand weeding once in each crop with RDF	8.4	8.5	8.3	174.9	164.8	162.0	23.8	22.6	23.0	115.4	117.3	106.3
Butachlor followed by 2,4-D in each crop with RDF	8.2	8.4	8.2	172.4	161.3	174.9	23.0	21.7	23.2	100.6	99.5	107.9
Butachlor followed by 2,4-D in each crop with RDF (25 % N substitution through organic manure)	9.2	9.3	9.4	190.1	189.5	192.8	25.7	24.9	25.4	108.6	110.2	99.1
Butachlor (autumn) or pretilachlor (winter) followed by 2,4-D in each crop with RDF	8.4	8.7	8.3	177.4	165.7	175.9	22.9	21.6	23.1	106.4	115.1	101.3
Butachlor (autumn) and pretilachlor (winter) followed by 2,4-D in each crop with RDF (25 % N substitution through organic manure)	9.4	9.8	9.4	200.3	188.6	195.0	26.3	24.8	26.2	112.0	114.1	108.7
CD $P=0.05$	0.9	1.0	1.1	20.7	18.8	23.7	2.3	NS	2.6	NS	NS	NS
CV (%)	8.7	9.2	10.0	9.3	8.9	10.9	8.5	10.8	8.9	7.6	9.1	8.0

20th – after harvest of 20th crop (winter rice 2011), 21st - after harvest of 21st crop (autumn rice 2012), 22nd – after harvest of 22nd crop (winter rice 2012)
 RDF= 40:20:20 and 60:20:40 kg ha^{-1} N:P₂O₅:K₂O in autumn rice and winter rice, respectively.

Table 5. Microbial population count ($-\log \times 10^{-6}$ cfu g^{-1} soil) and microbial biomass carbon ($\mu g g^{-1}$ of soil) in soil after harvest of 20th crop

Treatment	<i>Azospirillum</i>	<i>Azotobacter</i>	PSB*	MBC [#]
Hand weeding once in each crop with RDF	17.4	12.6	11.7	335.2
Butachlor followed by 2,4-D in each crop with RDF	14.2	9.4	8.6	328.3
Butachlor followed by 2,4-D in each crop with RDF (25 % N substitution through organic manure)	15.9	11.5	10.6	359.7
Butachlor (autumn) or pretilachlor (winter) followed by 2,4-D in each crop with RDF	14.6	9.9	9.4	337.0
Butachlor (autumn) and pretilachlor (winter) followed by 2,4-D in each crop with RDF (25 % N substitution through organic manure)	16.3	11.4	10.9	370.1
CD $P=0.05$	1.6	1.3	1.2	22.6
CV (%)	8.5	9.9	9.9	5.4

RDF= 40:20:20 and 60:20:40 kg ha⁻¹ N:P₂O₅:K₂O in autumn rice and winter rice, respectively

*PSB – phosphate solubilizing bacteria

[#]MBC – microbial biomass carbon

Table 6. Activity of acid phosphatase (μg p-nitrophenol g^{-1} dry soil h^{-1}), urease ($\mu g g^{-1}$ soil h^{-1}) and dehydrogenase (μg TPF g^{-1} dry soil 7days⁻¹) in soil after harvest of 20th crop

Treatment	Acid phosphatase	Urease	Dehydrogenase
Hand weeding once in each crop with RDF	200.4	341.3	106.0
Butachlor followed by 2,4-D in each crop with RDF	136.8	340.5	124.6
Butachlor followed by 2,4-D in each crop with RDF (25 % N substitution through organic manure)	170.2	369.5	145.5
Butachlor (autumn) or pretilachlor (winter) followed by 2,4-D in each crop with RDF	151.4	328.9	117.7
Butachlor (autumn) and pretilachlor (winter) followed by 2,4-D in each crop with RDF (25 % N substitution through organic manure)	184.6	366.8	141.2
CD $P=0.05$	19.3	24.0	14.7
CV (%)	9.5	5.7	9.6

RDF= 40:20:20 and 60:20:40 kg ha⁻¹ N:P₂O₅:K₂O in autumn rice and winter rice, respectively

herbicides significantly increased the dehydrogenase activity in soil while the effect of rotation of pre-emergence herbicide was non-significant, irrespective of organic manure addition. The dehydrogenase activity in soil increased significantly due to addition of organic manure for partial substitution of N-fertilizer. The highest dehydrogenase activity of 145.5 μg TPF g^{-1} dry soil 7 days⁻¹ was observed with organic manure addition

for partial N-fertilizer substitution without herbicide rotation.

Irrespective of the treatments, the population of *Azotobacter* was more than that of *Azospirillum*. The microbial biomass carbon in soil was not affected by application of herbicides. Application of herbicides significantly reduced the population of phosphate solubilizing bacteria in soils. The observation is in

affirmation to that reported by Latha and Gopal (2010). The decrease in acid phosphatase activity in soil with application of herbicide is in conformity to the findings of Wang *et al.* (2007), as the negative effect of butachlor on acid phosphatase is augmented in presence of cadmium. The urease activity was not affected by herbicide application compared to hand weeding, while addition of organic manure for partial N-fertilizer substitution significantly increased it over sole chemical fertilizer.

Conclusion

It may be concluded that application of herbicides with organic manure sustained yield of rice and fertility of soil over a decade. Future studies on herbicide-microorganism-nutrient interaction in the crop and weed plant rhizosphere would aid in better understanding of the long-term effect of weed and nutrient management practices.

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