

Effect of Herbicides and Nutrient Management on Soil Enzyme Activity

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Abstract

A field experiment was conducted at Student Farm, College of Agriculture, ANGRAU, Rajendranagar, Hyderabad, during Kharif 2010 to study the effect of two herbicides (Butachlor and Cyhalofop-butyl) and nutrient management practices on soil urease, phosphatase and dehydrogenase activity in a clay loam soil using rice as test crop. The effects of herbicides on soil enzyme activity showed that there was an increase in soil enzyme activity from 0 to 60 days after transplanting of the crop irrespective of the treatment of the soil. The increase with age of the crop, however, was less during the first two stages of the crop and with significant increase at later stages. The effect of nutrient management on enzyme activity showed that, urease, acid phosphatase and alkaline phosphatase activity was increased with number of days and attained maximum activity at 60 days after transplanting (DAT), thereafter the activity decreased gradually to original level at 105 DAT. The comparison of enzyme activity using CD of interaction between treatments for the incubation periods indicate that at 20 days after transplanting, NPK treatment recorded higher enzyme activity than control. At 60 DAT, all the enzymes exhibited the highest value for NPK treatment while the lowest activity was recorded for untreated control

Keywords: Herbicides, nutrient management, soil enzyme activity, rice.

The intensive development of farming systems has resulted in a large-scale application of crop protection chemicals. Weeds are the major biological constraint in most rice growing areas of the world. The lack of suitable weed control alternatives has led to increase in reliance on herbicides in many rice growing areas and their use is increasing as they are less expensive and convenient than manual labor, very effective and easy to use. Herbicides may affect non-target organisms including microorganisms (Latha and Gopal, 2010). This is especially true of rice

ecosystem which is composed of extremely large number of very diverse microbial sub-habitat in space and time. Hence, a field experiment was conducted to study the effect of two herbicides (Butachlor and Cyhalofop-butyl) and nutrient management practices on soil enzyme activity in a clay loam soil using rice as test crop.

Materials and Methods

The field experiment was conducted at Students Farm, College of Agriculture, ANGRAU, Rajendranagar, Hyderabad, during Kharif 2010. The rhizosphere soils of an ongoing experiment from AICRP on weed control were selected and the effect of herbicides, weed management practices and nutrient management practices on soil urease, phosphomonoesterases (acid and alkaline) and dehydrogenase activity has been carried out with rice (variety –MTU1010) as the test crop. The soils taken from AICRP weed control plots in Students Farm, where the fertilizer treatment imposed as subplots in herbicide treated plots, were analyzed using standard procedure. Urease in soil was determined as described by Tabatabai and Bremnar (1972). Procedure of Tabatabai and Bremnar (1969) and Eivazi and Tabatabai (1977) were adopted for assay of acid and alkaline phosphatase, respectively and dehydrogenase activity was determined as described by Casida *et al.* (1964). Calculated quantities of fertilizers and herbicides were applied in each plot based on recommended dose. The recommended fertilizer dose of 150:60:60 NPK was applied, with nitrogen in three split doses viz; basal, active tillering and panicle initiation stages. Phosphorous was applied completely as basal application and potassium in two splits, at basal and at panicle initiation stages. Butachlor @ 1kg ai ha⁻¹ was applied three days after transplanting (DAT) and cyhalofop-butyl @ 1kg ai ha⁻¹ at 20 DAT. Farmers practice of hand weeding was also done at 20 and 40 DAS. The experiment was conducted in a randomized block design with three replications. Initial soil sample was collected immediately after planting and subsequent

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soil samples were collected at fifteen days interval till the crop harvest. The moisture status was maintained by irrigating the field whenever necessary. All the data on enzyme activity was analyzed using two-way ANOVA described by Snedecor and Cochran (1967).

Results and Discussion

The activity of urease (expressed as μg of NH_4^+ released g^{-1} soil h^{-1}) as influenced by the herbicide treatments without RDF (control) is presented in Table (1). A close perusal of data indicates that significant difference exists between herbicide treatments and periods of study and their interactions at 5% level. Urease activity in control increased from 0 DAT (1.9) to 60 DAT (3.9). Thereafter, the activity decreased to almost 0 day level at 105 DAT. Using the CD of interactions (treatments and DAT) to test the significance of the differences in the given treatments, it can be seen that urease activity levels in the herbicide treated plots are significantly higher than control for all the periods except 0 and 105 DAT.

At 45 DAT, Butachlor showed significantly higher activity of 6.9 followed by Cyhalofop-butyl (5.6), farmer's practice (3.9) and control (3.8). At 60 DAT, Butachlor recorded the highest activity (8.1) followed by Cyhalofop-butyl (7.8), farmer's practice (4.5) and control (3.9).

The activity of urease (expressed as μg of NH_4^+ released g^{-1} soil h^{-1}) as influenced by the herbicide treatments plus RDF is presented in Table (2). The urease activity was found to be higher in nutrient applied plots compared to control. This increased urease activity may be attributed to increased soil nutrients used by urease enzyme releasing microorganisms. Significant difference exists between herbicide treatments and periods of study and their interactions at 5% level. Urease activity in control increased from 0 DAT (2.8) to 60 DAT (7.5). Thereafter, the activity decreased to almost 0 day level at 105 DAT. It was observed that urease activity levels in the herbicide treated plots are significantly higher than control for all the periods except 0 and 105 DAT. At 45 DAT, Butachlor shows significantly higher activity of 9.5 followed by Cyhalofop-butyl (7.2), farmer's practice (6.7) and control (5.6). At 60 DAT, Butachlor recorded the highest activity (12.1) followed by Cyhalofop-butyl (9.6), farmer's practice (9.3) and control (7.5). Similar trend was followed for Phosphomonoesterase and Dehydrogenase activity also.

The two herbicides treatment along with or without RDF has found to have a stimulatory effect on acid phosphatase activity in the present study (Table 3). Highest activity was observed in the treatment with NPK application as well herbicides applications i.e. RDF + herbicides followed by control (Table 4). This increase in acid phosphatase activity might be due to increase in P- solubilizer population due to the degradation of applied herbicides or products might have served as a carbon source. Similar trend is followed for alkaline phosphatase activity also. The activity of alkaline phosphatase (expressed as μg of 4-nitrophenol released g^{-1} soil h^{-1}) as influenced by the herbicide treatments without fertilizer application is presented in Table (5) and with fertilizer application is given in Table (6). Treatment receiving RDF + herbicides showed stimulatory effect on soil phosphatase enzyme activity which may be attributed to use of applied fertilizers as a nutrient source by soil microorganisms releasing phosphatase enzymes in the soil. Effect of herbicides on dehydrogenase activity in flooded rice soil also showed similar trends (Table 7 and 8). This is in agreement with works of Reddy (1997) who reported highest dehydrogenase, urease and phosphatase activity with RDN + FYM compared to treatment with only inorganic source of RDF. Similarly, Lalfakzuala *et al.* (2006) reported that fertilizer treatment increases microbial population number and microbial enzymatic activity. Kondratowicz (2007) also found that fertilization with nitrogen and manure resulted in an increase in microbial populations and a higher enzymatic activity in soil.

Since herbicides are used when the crop is either absent as pre-emergence or at its early stage of growth as post-emergence, a high proportion of herbicide reaches the soil and accumulates in the microbiologically active top layer of 0 to 15cm of soil. The detracting effect of herbicides towards all bacteria, fungi and enzyme activities decreased with time. This is because of the recovery of microbial population and enzyme activities after initial inhibition due to microbial adaptation to these chemicals or due to their degradation. It can also be due to microbial multiplication on increased supply of nutrients available in form of microorganisms killed by herbicides (Latha and Gopal, 2010). They studied the effect of herbicides 2, 4- D, butachlor, pretilachlor and pyrazosulfurol on soil enzyme activities. Among these herbicides tested, the enzyme activity inhibition followed a trend, butachlor > 2, 4-

D > pretilachlor > pyrazosulfuron. Kavitha *et al.* (2011) found that application of herbicide generally disturb and alter the biological equilibrium in the soil, so at the initial stages, that is, 20 days after transplanting of rice, the pre-emergent application of Pretilachlor @ 0.75 Kg ai ha⁻¹ lower the microbial population and hence enzyme activity. But at later stages, enzyme activity and microbial population was increased. Perucci *et al.* (2000) studied the effect of sulfonylurea herbicides on soil enzyme activity. These herbicides were applied at field rates and 10 fold field rates. The higher rate of herbicide application impaired microbial parameters to a greater degree. It is found that these xenobiotic compounds force the soil microbial biomass to direct a large part of its energy budget into reducing mineralization activity. This situation had a long lasting negative effect on soil fertility.

Nutrient management practices like application of organic manures and mineral fertilization caused an increase in the abundance of soil microorganisms and enzymatic activity. Mineral fertilization resulted in the lowest urease activity presumably by the small amounts of organic residues left in the soil (Balezientene and Kilimas, 2009). The higher application of fertilizer lowers the pH value and affects the microbial activity due to induced soil acidity. This result showed that composition change in microbial community as affected by a fertilization effect. A balanced amount of NPK and organic manures will increase the enzyme activity (Joa, 2010).

Though in general, application of chemical fertilizers stimulated the growth and multiplication of microorganisms, increased dosage was found to inhibit the survival of microbe due to osmotic stress created by fertilizers (Bharathi *et al.* 2011). A significant increase in soil enzyme activity of urease (78µg of NH₄⁺ g⁻¹ day⁻¹), in unfertilized control plot was followed by 75% of RDF of NPK at 30 DAS. There is further reduction in enzyme activity when 150% of RDF of NPK was applied due to the negative impact of higher dose of chemical fertilizers alone on survival of microorganisms.

A study was conducted to find the effect of herbicides on dehydrogenase activity in flooded rice soil. The dehydrogenase activity in flooded rice soil increased up to 40 days after transplanting (DAT), after which the activity decreased with no significant difference at 120 DAT. The sharp increase in all treatment at 20 and 40 DAT represents the most

active growth period of rice crop and could be due to proliferation of anaerobic micro-flora in the rhizosphere. The stabilized activity of dehydrogenase at lower levels at 120 DAT could be due to the fact that soil attains moisture content between field capacity and permanent wilting point and represents the effect of soil drying on dehydrogenase activity. Also, the herbicides used at recommended dosage were non-inhibitory on dehydrogenase activity (Rao and Raman, 1998).

The effects of herbicides on soil enzyme activity showed that there was an increase in soil enzyme activity from 0 to 60 days after transplanting of the crop growth irrespective of the treatment of the soil. The increase with age of the crop, however, was less during the first two stages of the crop and with significant increase at later stages. The effect of nutrient management on enzyme activity showed that, urease, acid phosphatase, alkaline phosphatase and dehydrogenase activity was increased with number of days and attained maximum activity at 60 days after transplanting (DAT), thereafter the activity decreased gradually to original level at 105 DAT. The comparison of enzyme activity using CD of interaction between treatments for the incubation periods indicate that at 20 days after transplanting, NPK treatment recorded higher enzyme activity than control. At 60 DAT, all the enzymes exhibited the highest value for NPK treatment while the lowest activity was recorded for untreated control. It was found that soil enzyme activities were influenced by the system of agriculture, input of fertilizers and pesticides and decreases with depth. The effect of cultivation induced significant changes in the quality, chemical composition and molecular size of organic matter, which in turn influenced the activities of enzymes involved in C, N and P cycle.

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Table 1: Effect of Herbicides and Nutrient Management on soil urease (control)

Treatments	Urease Activity ($\mu\text{g of NH}_4^+ \text{ g}^{-1} \text{ soil h}^{-1}$)							
	Days after transplanting							
	0	15	30	45	60	75	90	105
Control	1.9	2	2.6	3.8	3.9	3.5	2.8	2.1
Cyhalofop-butyl	2.6	3.7	4.5	5.6	7.8	5.4	3.8	2.8
Butachlor	2.8	3.8	4.8	6.9	8.1	5.8	4.2	3.2
Farmer's Practice	2.2	2.3	2.7	3.9	4.5	4.1	3.8	2.5

Analysis of Variance	C.D.	SE(d)	SE(m)
Herbicides	0.108	0.053	0.037
Urease	0.153	0.075	0.053
Herbicides x Urease	0.305	0.149	0.105

Table 2: Effect of Herbicides and Nutrient Management on soil urease (RDF)

Treatments	Urease Activity ($\mu\text{g of NH}_4^+ \text{ g}^{-1} \text{ soil h}^{-1}$)							
	Days after transplanting							
	0	15	30	45	60	75	90	105
Control	2.8	3.2	4.9	5.6	7.5	5.2	4.8	2.7
Cyhalofop-butyl	3	3.7	5.1	7.2	9.6	7.4	5.2	3.3
Butachlor	3.8	4.2	7.3	9.5	12.1	9.1	6.9	3.9
Farmer's Practice	2.7	3.4	3.9	6.7	9.3	6.7	4.9	3.2

Analysis of Variance	C.D.	SE(d)	SE(m)
Herbicides	0.127	0.062	0.044
Urease	0.179	0.087	0.062
Herbicides x Urease	0.358	0.175	0.124

Table 3: Effect of Herbicides and Nutrient Management on soil acid phosphatase (control)

Treatments	Acid Phosphatase Activity ($\mu\text{g of 4-nitrophenol g}^{-1} \text{ soil h}^{-1}$)							
	Days after transplanting							
	0	15	30	45	60	75	90	105
Control	12.3	14.4	17.9	24.5	26.2	22.1	15.7	12.9
Cyhalofop-butyl	14.3	17.1	24.9	31.2	46.3	28.4	17.3	15.3
Butachlor	15.2	18.3	25.8	34.5	49.1	25.1	17.9	15
Farmer's Practice	13.7	14.5	20.5	29.8	34.8	23.6	16.1	13.9

Analysis of Variance	C.D.	SE(d)	SE(m)
Herbicides	0.118	0.057	0.041
Acid Phosphatase	0.166	0.081	0.057
Herbicides x Acid Phosphatase	0.332	0.162	0.115

Table 4: Effect of Herbicides and Nutrient Management on soil acid phosphatase (RDF)

Treatments	Acid Phosphatase Activity (μg of 4-nitrophenol g^{-1} soil h^{-1})							
	Days after transplanting							
	0	15	30	45	60	75	90	105
Control	15.5	20.1	43.1	50.2	62.8	53.2	40.7	20.2
Cyhalofop-butyl	20.5	48.8	70.7	100.8	130.7	70.8	43.3	24.3
Butachlor	18.8	25.4	54.7	69.7	107.1	62.7	44.9	23.3
Farmer's Practice	16.9	22.3	45.8	56.9	70.3	64.5	41.5	21.8

Analysis of Variance	C.D.	SE(d)	SE(m)
Herbicides	0.120	0.059	0.042
Acid Phosphatase	0.170	0.083	0.059
Herbicides x Acid Phosphatase	0.341	0.166	0.118

Table 5: Effect of Herbicides and Nutrient Management on soil alkaline phosphatase (control)

Treatments	Alkaline Phosphatase Activity (μg of 4-nitrophenol g^{-1} soil h^{-1})							
	Days after transplanting							
	0	15	30	45	60	75	90	105
Control	17.2	19.9	23.1	27.9	29.8	25.3	20.1	18.9
Cyhalofop-butyl	20.9	24.5	35.7	49.8	38.2	29.1	25.4	21.3
Butachlor	22.7	29.8	38.3	50.3	40.1	34.8	29.2	23.9
Farmer's Practice	18.4	20.9	26.9	34.1	36.4	27.1	22.3	18.7

Analysis of Variance	C.D.	SE(d)	SE(m)
Herbicides	0.121	0.059	0.042
Alkaline Phosphatase	0.172	0.084	0.059
Herbicides x Alkaline Phosphatase	0.343	0.167	0.118

Table 6: Effect of Herbicides and Nutrient Management on soil alkaline phosphatase (RDF)

Treatments	Alkaline Phosphatase Activity (μg of 4-nitrophenol g^{-1} soil h^{-1})							
	Days after transplanting							
	0	15	30	45	60	75	90	105
Control	20.7	31.9	49.7	60.4	73.4	62.6	56.2	22.5
Cyhalofop-butyl	21.5	40.4	70.9	80.7	110.5	88.7	60.3	30.5
Butachlor	22.9	46.9	79.8	105.8	106.4	100.7	65.4	34.9
Farmer's Practice	18.5	25.3	44.8	53.2	82.3	50.9	43.1	19.4

Analysis of Variance	C.D.	SE(d)	SE(m)
Herbicides	0.130	0.064	0.045
Alkaline Phosphatase	0.184	0.090	0.064
Herbicides x Alkaline Phosphatase	0.368	0.180	0.127

Table 7: Effect of Herbicides and Nutrient Management on dehydrogenase activity (control)

Treatments	Dehydrogenase activity(μg of TPF g^{-1} soil h^{-1})							
	Days after transplanting							
	0	15	30	45	60	75	90	105
Control	1.1	1.7	2.3	2.6	2	2.4	1.8	1.3
Cyhalofop-butyl	1.2	1.9	2.8	3	3.8	3.2	2.3	1.5
Butachlor	1.4	2.2	2.9	3.5	3.9	3.1	2.5	2.3
Farmer's Practice	1.3	1.9	2.6	2.7	3.1	2.5	2.2	1.5

Analysis of Variance	C.D.	SE(d)	SE(m)
Herbicides	0.127	0.062	0.044
Dehydrogenase	0.179	0.088	0.062
Herbicides x Dehydrogenase	0.359	0.175	0.124

Table 8: Effect of Herbicides and Nutrient Management on dehydrogenase activity (RDF)

Treatments	Dehydrogenase activity(μg of TPF g^{-1} soil h^{-1})							
	Days after transplanting							
	0	15	30	45	60	75	90	105
Control	1.3	1.7	2.7	3.1	4.1	3.8	3.1	1.8
Cyhalofop-butyl	1.6	1.9	3.3	4.2	4.5	4.1	3.3	2
Butachlor	1.7	2	3.5	4.7	4.9	4.4	3.5	2.2
Farmer's Practice	1.4	1.8	2.9	3.1	4.2	3.8	3.2	1.9

Analysis of Variance	C.D.	SE(d)	SE(m)
Herbicides	0.116	0.057	0.040
Dehydrogenase	0.164	0.080	0.057
Herbicides x Dehydrogenase	0.328	0.160	0.113

