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Short communication

Herbicides effect on soil enzyme dynamics in direct-seeded rice

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Dry direct-seeding of rice (DDSR) has emerged as a viable option to combat the rising production costs and shortage of water and labour. Dry sown or semi dry system of rice cultivation is a unique and extensively adopted system of rice cultivation in Kerala and it accounts for more than 60 per cent of the area under rice during *Kharif* season. However, weeds are the most serious biological constraint in dry-seeded rice because of the absence of stagnant water. Use of low dose, high efficiency herbicides is becoming more prevalent which remain more persistent in soil. This necessitates the need to assess the risk of herbicide persistence in soil on soil life. Soil enzyme activity is an indication of microbial activity, which is an integral part of soil environment. Herbicides are extraneous to soil component pools, and are expected to affect the catalytic efficiency and behavior of soil enzymes (Sannino and Gianfreda 2001), which contribute to total biological activity of the soil-plant environment under different situations. Dehydrogenase activity is commonly used as an indicator of biological activity in soils and this enzyme is considered to exist as an integral part of intact cells but does not accumulate extra cellular in soil. Urease enzyme is responsible for the hydrolysis of urea fertilizers applied to the soil into NH_3 and CO_2 with the concomitant rise in soil pH (Andrews *et al.* 1989). This, in turn, results in a rapid N loss to the atmosphere through NH_3 volatilization (Simpson *et al.* 1984). Due to this role, urease activities in soils have received a lot of attention since it was first reported, a process considered vital in the regulation of N supply to plants after urea fertilization.

Various studies have revealed that the herbicides can cause qualitative and quantitative change in enzyme activity (Xia *et al.* 2011). Bensulfuron-methyl, azimsulfuron and pyrazosulfuron-ethyl belongs to sulfonylureas are extensively used to control a wide range of weeds inhibiting acetolactate synthase, a key enzyme in protein synthesis of plants. However, the knowledge about the effect of herbicides on soil enzyme activities in dry sown rice has been limited. In this background, the present study was conducted to find out the effect of these herbicides on soil enzymes *viz*. dehydrogenase and urease for getting a better understanding of the possible response of soil microbial activities to low dose herbicides.

Field experiment was conducted in Vellayani located at 8.5°N latitude and 76.9 °E longitudes at an altitude of 29 m above MSL. A warm, humid, tropical climate is experienced by the experimental area. The soil of the experimental site belonged to textural class of sandy clay which the taxonomical order was oxisol. The soil pH was 5.41 and EC was normal, high in organic carbon, available P and medium in available N and K. Experiment was conducted during *Kharif* season of 2014 by dry direct-seeding in semi-dry system where the field was flooded after 45-50 DAS under randomised block design with three replications. The treatments included different combinations of pre-emergent herbicides with post-emergent herbicide and hand weeding. The new low dose herbicides used were bensulfuron-methyl + pretilachlor, pyrazosulfuron-ethyl (pre-emergent herbicides) and azimsulfuron (post-emergent herbicide) along with a traditional herbicide oxyflourfen applied as pre-emergent herbicide. Pre-emergent herbicides were applied one day after sowing on to the surface of soil using knapsack sprayer with flood jet nozzle while post emergent herbicides were applied at 25 DAS on to the weed flora. The herbicides were sprayed at recommended rates of 660 g/ha for bensulfuron-methyl + pretilachlor as per Sanjay *et al*. (2013), pyrazosulfuron 25 g/ha as per Latha and Gopal (2010) and azimsulfuron 30 g/ha as per Jayadeva *et al.* (2011). The soil samples were taken at a depth of 15 cm before and at 15, 30, 45 and 60 days after sowing (DAS) for determining dehydrogenase. Urease enzyme activity samples were taken at 15 and 30 DAS. Dehydrogenase activity was measured following reduction of 2,3,5-triphenyltetrazolium chloride (TTC) to red-coloured triphenylformazon (TPF), which were determined spectrophoto

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metrically (Casida *et al*. 1964). Urease activity of herbicides treated as well as control soil was determined by titration method described by Broadbent *et al.* (1964).

There was variation in soil enzyme activity in soils as influenced by different herbicide treatments at different period of time. It was observed that dehydrogenase enzyme activity followed an asymptotic behaviour. This pattern could be described by an increase at initial stages reaching a maximum at active tillering stage of the crop followed by a decline towards the harvest stage. At 45 DAS, it showed an increase and after that at 60 DAS, a declining trend was seen. The dehydrogenase activity was significantly higher at 45 DAS compared to other time of plant growth, *viz.* 15, 30 and 60 DAS, which might be due to the spurt in microbial population with the addition of exudates or rhizodeposition during the growth stages *i.e*. up to 45 days. Since dehydrogenase is an endo enzyme, the activity is maximum at the active growth stage of plant (Lu-sheng *et al.* 2005). The highest activity of dehydrogenase recorded at 45 DAS irrespective of the treatment may be due to the fact that it corresponds to the active growth stage of the crop and there is enhanced rhizosphere activity coupled with commencement of flooding from 40 DAS in semi dry rice. These results corroborate the findings of Vandana *et al.* (2012) who reported increase in soil dehydrogenase enzyme activity after herbicide application from 7^{th} to 28^{th} day of incubation which was due to increase in microbial community composition with the capability to utilize herbicide as carbon source.

The data suggested that the detracting effects of soil applied pre-emergent herbicides on soil enzyme activity decreased with time. At 45 DAS, pyrazosulfuron-ethyl 25 g/ha as pre-emergence showed significantly higher activity followed by pyrazosulfuron-ethyl *fb* hand weeding and at 60 DAS, pyrazosulfuron-ethyl with hand weeding recorded the highest activity followed by oxyfluorfen and hand weeding. Among pre-emergent herbicides, oxyflourfen treated plots recorded the highest dehydrogenase activity at 15 days after application when compared to control (weedy check) along with pyrazosulfuron-ethyl. This is contradictory to the findings of Baboo *et al.* (2013) who reported lowest soil dehydrogenase activity in soil treated with the same herbicide under *invitro* situation which might be due to difference in soil biota under field situations. Having the highest dehydrogenase activity at 15 DAS, oxyflourfen could be recommended as a safe herbicide for dry sowing. Hence it could be inferred that herbicides used at recommended rates were non inhibitory on dehydrogenase activity in dry sown situations.

The activity of urease (expressed as urea hydrolysed per gram of soil per hr) as influenced by the herbicide treatments was not inhibited by any of the herbicide treatments. Higher values of urease activity were recorded at both 15 and 30 DAS and higher urease enzyme activity might be attributed to the higher amount of substrate availability during these stages. The urease activity was found to be higher in nutrient applied as well as weedy check plots. This increased urease activity may be attributed

Table 1. Dehydrogenase enzyme activity (TPF/g of soil/ 24 hr) and urease activity (urea hydrolysed/g of soil/hr) as influenced by herbicide treatments

Treatment	Dehydrogenase enzyme activity (TPF /g of soil /24 hr)				Urease activity (Urea hydrolyzed/g^{-}of soil/hr)	
	15 DAS	30 DAS	45 DAS	60 DAS	15 DAS	30 DAS
Bensulfuronm-ethyl + pretilachlor 60 + 600 g/ha	129.6	182.6	205.8	198.7	70.8	74.4
Bensulfuronm-ethyl + pretilachlor 60 + 600 g/ha + hand weeding at 40 DAS	136.2	179.8	354.7	282.3	67.4	60.0
Bensulfuronm-ethyl + pretilachlor 60 + 600 g/ha + azimsulfuron 30 g/ha as (post-emergence)	150.1	234.0	233.0	127.1	66.0	67.2
Pyrazosulfuron-ethyl 25 g/ha as (pre-emergence)	173.0	247.0	567.8	256.6	90.6	64.5
Pyrazosulfuron-ethyl 25 g/ha as (pre-emergence) + hand weeding at 40 DAS	174.1	131.7	540.7	370.2	58.5	77.0
Pyrazosulfuron-ethyl 25 g/ha as (pre-emergence) + azimsulfuron 30 g / ha	180.1	83.5	159.9	242.2	78.3	75.4
Oxyfluorfen 0.15 kg/ha	275.5	155.9	230.4	295.8	71.3	78.6
Oxyfluorfen 0.15 kg/ha + hand weeding at 40 DAS	239.9	123.7	429.0	336.5	72.6	70.4
Oxyfluorfen 0.15 kg/ha + azimsulfuron 30 g /ha	290.9	99.4	236.5	316.1	84.4	62.1
Hand weeding at 20 and 40 DAS	229.6	177.4	326.5	313.3	80.6	50.4
Weedy check	169.0	235.8	410.3	294.9	80.5	83.0
LSD (P=0.05)	5.43	30.5	9.27	9.86	5.72	7.59

to increased soil nutrients used by urease enzyme releasing microorganisms. (Vandana *et al*. 2012). Hence it could be inferred that herbicides either used as pre-emergence or post-emergence remains in the soil and cause alternations in soil enzyme activities with respect to different days after treatments.

SUMMARY

A field experiment was conducted to study the effect of new generation herbicides on soil enzymes, *viz*. dehydrogenase and urease for getting a better understanding of the possible response of soil microbial activities to low dose herbicides in dry sown rice. The treatments included different combinations of pre-emergent herbicides (bensulfuron-methyl + pretilachlor, pyrazosulfuron-ethyl) with post-emergent herbicide (azimsulfuron) and hand weeding along with a traditional herbicide oxyfluorfen. The effect of herbicides on soil enzyme dynamics showed that there was an increase in soil dehydrogenase enzyme activity (expressed in triphenylformazanhydrolysed /g of soil /24 hr) from 15 to 45 DAS after an initial decline. The activity of urease (expressed as urea hydrolysed per gram of soil per hr) as influenced by the herbicide treatments was not inhibited by any of the herbicide treatments. Hence it could be inferred that herbicides either used as pre-emergence or post-emergence remains in the active top soil and cause alternations in soil enzyme activities with respect to different days after treatments. The treated herbicides affected the soil enzyme activity but none of them registered highly negative effect on any of the soil enzyme activity in the present study under dry sowing.

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